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# Cultivating Clean Energy

## The Promise of Algae Biofuels

### Author

Catie Ryan, *Terrapin Bright Green, LLC*

### Lead Editor

Alice Hartley, *Terrapin Bright Green, LLC*

### Project Managers

Bill Browning, *Terrapin Bright Green, LLC*

Chris Garvin, *Terrapin Bright Green, LLC*

Nathanael Greene, *Natural Resources Defense Council*

Cai Steger, *Natural Resources Defense Council*



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### NRDC Algae Biofuels Advisory Committee

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Walter Adey	Curator of Algae, The Smithsonian Institute
Al Darzens	Applied Science, Biomass Research, National Renewable Energy Laboratory (NREL)
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Arthur Grossman	Department of Plant Biology, The Carnegie Institution
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Production: Tanja Bos, [tanja@bospoint.com](mailto:tanja@bospoint.com)

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# Introductory Letter

**A**lgae-derived gasoline, diesel, and jet fuel sound like the imaginings of science fiction, but a growing number of entrepreneurs, investors, academics, and policy makers are trying to make them reality. The economic, national security, and environmental costs of our dependence on oil become clearer every day, and algae may be able to provide large quantities of locally produced biofuels with minimal environmental impacts.

The algae biofuels industry is comprised of many pathways to produce fuels from algae and is developing rapidly, with most companies operating in “stealth” mode. This makes measuring progress toward the promise of algae extremely difficult. Given the thousands of different algal strains, multitude of cultivation and harvest methods, wide range of algae products, and host of technologies used to convert these products into different transportation fuels, algae-based biofuels make up a broad and difficult-to-categorize family of technologies and production pathways. Furthermore, algal technologies are evolving rapidly. Worldwide investments in algae were \$32 million in 2007 and surpassed the \$300-million-dollar mark in 2008; Sapphire Energy led with a \$100 million dollar investment in research and development from Bill Gates.<sup>1,2</sup> Today, more than 50 companies have received funding to focus on algae fuels.<sup>3</sup>

In the face of this sprawling, dynamic industry, it is hard to develop a clear picture of the environmental pros and cons of a full system to grow algae and turn it into fuels. Many developers and researchers in the industry are studying or analyzing single processes within a stage of biofuel production—cultivating algae, drying it, or separating it into its useful products—and thus may possess only a partial picture of the environmental impacts upstream and downstream from their respective focus area.

In this regard, it is perhaps instructive to consider the economic, political, and technological challenges currently faced by first-



generation biofuel technologies such as corn ethanol and palm oil biodiesel. Had the environmental impacts of these technologies been better anticipated and managed, the fuel industry might not be considering algae-derived biofuels at all. Given that algae are being explored as a third-generation fuel source, we must now consider how to be smart about the types of effects that were previously ignored.

To properly assess algae biofuels, we need a way to see the big picture—to develop the full life cycle of algae-to-biofuel production and analyze all potential impacts. This is especially important for a technology that we hope will contribute on a meaningful scale to meeting our transportation energy needs.

This report provides an overview of the potential positive and negative environmental externalities of algae biofuel processes and technologies. In doing so, we hope to provide a methodology and logic that can be used in the future to analyze all inputs and outputs associated with every potential process in an algae-to-biofuel production pathway. Specifically, the report identifies key environmental issues to be considered across all stages of an algae biofuel production; proposes a mapping framework for these algae-to-biofuel pathways; summarizes what is known and unknown about the potential environmental impacts of each algae-to-biofuel process; and identifies areas of future research need and recommends policy and industry actions to improve the environmental sustainability of the industry and its fuel production practices.

We do not predict which approach to algae biofuels will ultimately succeed or which will be preeminent from an environmental perspective. Information currently available on the full life cycle of any algae-to-biofuel production pathway is insufficient to make useful quantitative comparisons among algae-based biofuel (or other first- or second-generation biofuel) production pathways. The variety of technologies and production processes under development is wide-ranging and difficult to classify, and we have not tried to describe them all.

Instead, we offer a basis for consensus in considering a full-system, life cycle approach in assessing the sustainability of relevant technologies. We also hope that stakeholders can work together to clarify the unknown environmental impacts and consider how best to maximize the environmental benefits from algae biofuels, minimize the risks, and avoid unintended consequences.

— Nathanael Greene and Cai Steger  
*Natural Resources Defense Council*

# Executive Summary

**T**ransportation emissions are among the largest sources of global warming pollution in the United States. But America cannot continue to rely on dirty fuels to power our nation—we need clean, renewable energy sources that will help advance our clean energy economy. Efforts to curb global warming, concerns about depleting petroleum reserves, and national security issues have led scientists, industry, and governments to investigate new energy sources. One of the most promising new sources of sustainable fuel is algae-based biofuels.

## **We Need Sustainable Fuels That Can Meet our Fuel-Consumption Needs**

Liquid fuel production alternatives have focused on ethanol from agricultural yields, biodiesel from cellulosic plant materials, and hydrogen production. Unfortunately, each technology brings with it a host of challenges, such as increased food prices because of competition with agriculture-based fuels crops, challenges to converting cellulosic materials, and outdated infrastructure unfit to transport and store hydrogen. One of the greatest obstacles facing the biofuels industry is supplying enough sustainable feedstock to produce the quantities of fuel needed at affordable prices.

Today, scientists are converting the lipids and hydrocarbons produced by algae into a variety of fuels and these algae-based biofuels are being touted by some as a path to a sustainable energy supply.

However, understanding issues of “scale” is vital to the production of sustainable biofuels. In the United States alone, 140 billion gallons of motor

fuel are consumed every year;<sup>4</sup> worldwide, that number reaches more than 320 billion.<sup>5</sup> At this scale, what seem like minor environmental impacts when producing one gallon of biofuels can quickly escalate into an industry liability and environmental disaster. Although the water demands for producing a gallon of algae-based biofuel may seem acceptable, on the scale of tens of billions of gallons over the course of a decade, scaled water demands may have a catastrophic effect on regional watersheds from which water is extracted and released. This notion of scale can be applied to key environmental issues—water, land, soil and biodiversity, air, and energy—and remains an overarching concern with any fuel production pathway. Identifying, managing, and mitigating the core environmental impacts associated with algae biofuels production is the first step in supporting the development of a sustainable biofuels industry.

## Production Pathways for Algae-Based Biofuels and Associated Environmental Issues

Production consists of four primary processes: 1) algae cultivation, 2) biomass harvesting, 3) algal oil extraction, and 4) oil and residue conversion. This report explores each production process and its associated environmental implications as well as their relationships to one another in terms of a collective production pathway. Understanding the relationship of algae-to-biofuel production to environmental issues, such as water resource management, land use impact, and energy balance, and identifying areas of unknown environmental impact, will help direct decision-making that can ensure a truly sustainable biofuel industry.

The four primary processes of algae-to-biofuel production are examined in two ways. The first part of this report conceptualizes production processes in a mapping framework to create a picture of the primary inputs and outputs that support five existing or potential production pathways. The second part explores these processes in terms of their system characteristics and potential environmental impacts.

## Pathways for Algae Cultivation

The purpose of algae cultivation is to grow raw algal biomass for the downstream production of fuel, based on the oil and residual components found in the biomass. In order to grow, algae need a source of water and essential nutrients, which are collectively referred to as the culture medium; algae cultivation facilities need land or area to occupy; and in most cases, algae need light to drive photosynthesis. The way in which water, nutrients, land, and light are supplied and managed for cultivation will affect the environment, especially at commercial scale.

There are many environmental issues with which sustainable algae cultivation will likely be challenged and impacts could vary drastically from system to system. At a minimum, the criteria for sustainable cultivation should consider impact of water and land usage and potential genetically modified organism (GMO) effects on biodiversity and ecosystem health, as well as the environmental impacts of infrastructure fabrication, installation, materials toxicity, electricity demands, and waste treatment. A key relationship is

that although water usage could be high, algae could biologically treat a contaminated water source.

## Pathways for Biomass Harvesting

Cultivation yields unprocessed algal biomass, which consists of algal cells suspended in the culture medium. Before algal cells can be separated into oil and residues for fuel and coproduct production, the raw biomass must be harvested from the culture medium. Harvesting consists of biomass recovery, which removes wet biomass from the cultivation system, and is often paired with dewatering and drying processes.

There are several techniques for recovering algal biomass, the implementation of which may vary depending on the cultivation system. Commonly used techniques discussed in this report include flocculation, dissolved air flotation, centrifugation, microfiltration, and decantation. Most recovery techniques require the chemical or mechanical manipulation of the culture medium that ultimately separates the biomass (product) from the process wastewater (output). The application of chemical additives can negatively affect the toxicity of the biomass and output water.

With the exception of heterotrophically cultivated biomass, algal biomass typically has high water content, and in most cases is not suited for conversion to biofuel products until it has undergone some degree of dewatering and/or drying. Dewatering and drying decrease the moisture content of the biomass to an acceptable level for the desired downstream conversion pathway(s). Dewatering decreases the moisture content of the biomass by draining or mechanical means. Drying continues this process by using a drum dryer, freeze dryer, spray dryer, and rotary dryer, or by solar drying.

Harvesting technologies are discussed briefly—in terms of recovery, dewatering, and drying—and several environmental benefits, concerns, and unknown impacts are identified. At scale, some of these drying systems are highly energy intensive and can affect the energy balance of production. At a minimum, the criteria for sustainable biomass harvesting should consider potential environmental toxicity of chemical additives and management of output water from recovery techniques and implications of energy-intensive drying techniques.

## Pathways for Algal Oil Extraction

In the oil extraction process, harvested biomass undergoes chemical or mechanical manipulation to isolate the algal oil from the cell membrane. TAG lipids (triglycerides) found in the algal cells are the primary product sought after for the purpose of biodiesel production. The remaining components of algal biomass (carbohydrates, proteins, nutrients, and ash) are referred to collectively as algal residue.

Algal oil extraction can be achieved via a number of techniques such as mechanical expulsion, solvent extraction, or supercritical fluid extraction. Osmotic shock and sonication are also discussed briefly to exhibit a range of pathways.

The criteria for sustainable oil extraction should consider energy inputs and potential environmental toxicity and safety concerns of chemical solvents, which have properties known to harm biological systems. Some impacts may have been unintentionally overlooked because of the limited availability of information about chemical and energy inputs to algal oil extraction techniques.

## Pathways for Oil and Residue Conversion

Algal biomass that undergoes oil extraction yields algal oil and residue, whereas biomass that is pretreated thermochemically (by pyrolysis and liquefaction) yields bio-oil and residue. The two oils, algal oil and bio-oil, are chemically distinct and must therefore be refined or “upgraded” under different conditions. Once the biomass is separated into oil and residue, transesterification can convert algal oil to biodiesel; hydroprocessing can convert algal oil and bio-oil to green or renewable biofuels; and much of the residue can be biochemically or thermochemically converted to a gaseous fuel or a solid, nutrient-rich coproduct such as animal feed.

Oil and residue conversion pathways include transesterification, fermentation, anaerobic digestion, gasification, pyrolysis, liquefaction, and hydroprocessing. These conversion processes are not unique to algal biomass and have been employed for use in conventional biofuel refining for some time. Nevertheless, a few environmental benefits, concerns, and unknown impacts can be identified. Some conversion processes are particularly energy intensive and output low-value

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*Our primary objective is to start the conversation about the life cycle environmental impacts of transforming algae into fuel. We hope that this conversation is carried forward, debated, and expanded upon to a much greater extent among stakeholders within the nascent algae biofuel industry.*

coproducts or byproducts with certain or potential environmental impacts. The criteria for sustainable conversion should consider potential energy demand and variety and usability of nonfuel products.

## Conclusions and Recommendations: Algae Biofuels Offer Promise, But Environmental Considerations Must be Addressed

Future implementation of algae biofuel production will need to address core environmental impacts in order to develop and commercialize a sustainable product. In the near term, industry may need to embrace the environmental benefits of biological services and nonfuel coproducts to make algae biofuels economically viable. Yet to develop a sustainable algae-to-biofuel industry, the environmental relationship among production processes needs to first be understood before any one process or pathway is championed over another. Over time, life cycle analyses and other tools for measurement may inform us that the primary environmental impacts of a production pathway are determined by select production processes.

Whereas the degree of impact from one pathway to another cannot be determined without a more thorough analysis, it can be foreseen that any two given pathways could have very different effects, both



*By proactively engaging in complex environmental analysis and life cycle calculations, stakeholders can help increase the odds of the long-term growth and successful development of algae-based biofuels and ensure the industry receives full consideration as one solution to our energy and environmental needs.*

anticipated and unknown. By assessing the entire production pathway, and linkage between individual processes, inputs, and outputs, many relationships have been identified between those processes and the environment. These relationships are summarized in terms of the environmental benefits, concerns, and unknown impacts of commercial-scale algae-to-biofuel production on water, land, soil and biodiversity, air, and energy. Of these five environmental issues, energy and water usage appear to have the greatest potential variable impact on commercial-scale production. Energy and water represent significant inputs or outputs in most production processes and their respective quantities and effects could vary dramatically from one pathway to another. The way water, nutrients, land, light, and other inputs are supplied and managed could have a significant effect on both the energy balance of a production pathway and the persistence of environmental quality.

Identified areas of unknown environmental impact highlight the need for both production data about system demands on water, land use, and energy, and a greater awareness of the long-term impacts of direct and indirect process inputs. Many of these unknown impacts are due simply to a lack of observable data, limited by the number of pilot- and commercial-scale projects.

Most of the work on using algae fuels as a clean energy source is focused on research and development and small demonstration systems; however, several companies are pursuing production-scale operations and testing their products for commercial use. Environmental challenges will persist in the production of algae biofuels until sustainable production processes are fully established. The need to improve the industry's environmental sustainability suggests a number of potential next steps for policy and industry leaders.

There are trade-offs between production pathways that are economically feasible and those that are environmentally sustainable, especially with respect to algae cultivation and biomass harvesting. From an economic standpoint, productivity of biomass and production of lipids must be high enough to bring the maintenance, energy, and scale-up costs down for the final fuel product to be at a practical level for the consumer, and finding the balance between economics and environmental impacts should not be oversimplified. Until major biological and technical barriers are overcome, the industry may need to engage otherwise underutilized land, water, and nutrient resources, biological services, and/or nonfuel coproducts to make algae biofuels economically viable. In order to develop and commercialize a sustainable product in the long term, research and development of algae biofuel production will need to closely address the externalities of scaled biofuel production processes—with respect to both direct and indirect land, water and energy inputs, chemical usage, land transformation, and materials fabrication and toxicity—and their potential impacts on the environment.

From a regulatory and policy standpoint, there are several tasks that could help push commercialization of the algae biofuel industry in a sustainable direction. At a minimum, the following actions should be undertaken:

- ▶ Clarify roles and responsibilities within government agencies
- ▶ Encourage subindustry collaboration
- ▶ Begin life cycle analysis (LCA) at the fuel product design phase
- ▶ Develop a regulatory roadmap
- ▶ Inventory all regulations and guidelines to establish an information resource

- ▶ Specify sustainability metrics and industry standards
- ▶ Adopt international standards for sustainable biofuels

More research is also needed on issues that will have environmental implications for algae biofuel processes. Industry should proactively address the following issues for their respective technologies, keeping in mind the relationships they maintain with other processes of algae-to-biofuel production:

- ▶ Conduct technoeconomic analyses
- ▶ Conduct a water balance
- ▶ Conduct energy and carbon balances
- ▶ Consider environmental impacts to native habitats in proximity to production and processing facilities; adopt low-impact development, operations, and maintenance practices
- ▶ Perform chemical recovery and use nonchemical substitutes for biomass recovery
- ▶ Consider materials toxicity and resource consumption for materials fabrication
- ▶ Begin life cycle analysis (LCA) at the fuel product design phase
- ▶ Encourage transparency of process inputs and outputs
- ▶ Improve understanding of how relationships between production processes define resource consumption and management (e.g., relationship between water inputs in one process and heat or electric energy inputs in same or other downstream process)

Establishing sustainable, scaled production pathways will require policy makers and the algae biofuel industry to leverage the environmental benefits of algae use and address the concerns and unknown impacts caused by unsustainable practices. Measurement of process inputs and outputs will be one of the keys to determining pathway sustainability, and collaboration among subindustries will help focus and unite such efforts.

The environmental benefits provided by algae-to-biofuel production have the potential to make significant contributions to a sustainable biofuels industry. However, associated environmental concerns and unknown impacts must be addressed through relevant technologies and policies to ensure the algae biofuel industry scales up in a consistent and beneficial manner.

## CHAPTER 1

# Potential Environmental Impacts of Algae-to-Biofuel Production

To characterize the relationship between the production of algae-based biofuels and environmental sustainability, this report identifies four core areas of potential environmental interest—water, land, soil and biodiversity—and analyzes their vulnerability to degradation or unsustainable usage from commercial-scale algae biofuel production processes. The report also puts forth observations concerning the potential carbon and energy balance of algae-based biofuels production.

### **Water**

Major components of regional water management concern water usage, downstream water, water quality, and groundwater and aquifer infiltration, each of which could be affected (positively or negatively) by the externalities of the various algae-to-biofuel production processes.

If algae biofuels demonstrate efficient water use and can economically treat wastewater, while maintaining downstream water quality and minimizing inhibitions to groundwater and aquifer infiltration, then algae may prove a valuable fuel feedstock. Absent these developments, algae biofuel production may not be commercially scalable, especially in water-constrained regions. Important to note, however, is that the effect of algae-to-biofuel production on regional water sources is not yet fully

understood and early emphasis by the algae industry on its water impact could mitigate many of these potential concerns.

### *Water Usage*

Water usage in algae cultivation promises to be a controversial calculation, with significant potential environmental impact. Preliminary calculations by algae scientists have raised the prospect that immense quantities of water could be needed to produce algae-based biofuel, but these early studies have not been included in this report. Without peer-reviewed science and commercial-scale algae biofuel production facilities, it would be premature to offer an explicit quantification of expected water usage. Nevertheless, it is reasonable to expect that how water is managed and/or recycled will profoundly influence the scalability



and the sustainability of commercial algae-to-biofuel production.

One factor that could limit the water consumption of the cultivation process is that algae can thrive in nutrient-rich eutrophic or mixed waters (such as with animal litter, tertiary wastewater, and agricultural or industrial effluents), the utilization of which would limit the impact on fresh water supplies. Further, algae naturally uptake nutrients, metals, and other pathogens from the water sources in which they grow, while also releasing or “injecting” oxygen back into that water. In doing so, algae essentially provide a biological method of treatment for municipal wastewater, industry effluents, eutrophic waterbodies, and other waste streams, potentially reducing the public cost burden of wastewater treatment.

Additionally, water rights and the local price tag on water will also affect system feasibility. In locations where water is limited, fresh water may come at a high cost, which could distort the economics of algae fuel production. Governance of water rights may also come into play, as the water resources and expanses of land needed to implement commercial-scale cultivation will likely cross property and administrative boundaries, which could increase transaction costs associated with development.

### ***Downstream Water***

Systems discharge is typically the most highly regulated and possibly toxic component of any industrial process because it releases process waste into the environment. This is a concern during the algae harvesting and processing stages, both of which can use potentially toxic chemicals. Regulations require that any discharge into a waterway must remove primary toxins and reduce the temperature of the waste stream to minimize damage to aquatic ecosystems. Nevertheless, downstream waterways have been notorious victims of industrial waste discharge. Industry pollutants infiltrate the soil and adjacent groundwater and can create dead zones in waterways where no life can exist.

### ***Water Quality***

The release or uptake of nutrients or organic solids from algae cultivation, harvesting, and processing has the potential to affect water quality. Water quality measures the suitability of water for a particular use based on specific chemical, physical, and biological

characteristics. Because of the lack of understanding about how industrial-scale algae production would interact with its natural environment, developing a complete picture of water quality impact from algae biofuels will take time.

As mentioned previously, there is some expectation that production processes involving chemical additives, especially within the harvesting process, will have the largest likely impact on downstream water quality. Conversely, some algae systems are designed specifically to clean up polluted water. Depending on how algae-based biofuel production is managed, especially with respect to algae cultivation and biomass harvesting processes, there is the potential to either mitigate or exacerbate wastewater and downstream water conditions.

However, by eliminating the need for agriculturally based biofuels, algae-derived biofuels could significantly, albeit indirectly, improve water quality. Nitrogen-rich fertilizers applied to cropland (including crops grown for biofuels) to increase yields are dissolved easily in rainwater or snowmelt runoff that flows to rivers, lakes, and oceans. Eroded soil and sediment can also transport considerable amounts of nutrients (e.g., organic nitrogen and phosphorus) and pesticides to waterbodies. By some estimates, as much as 50 percent of nitrogen applied as fertilizer finds its way to surface and groundwater.<sup>6</sup> Excess nutrients, especially of nitrogen and phosphorus, can dramatically change the structure and functioning of an ecosystem.<sup>7</sup> Nitrogen-driven bacterial growth (resulting from the decomposition of algal blooms) depletes water oxygen to the point that all higher organisms (plants, fish, etc.) die off.

### ***Groundwater and Aquifer Infiltration***

Depending upon the production processes used, algae-based biofuel production could limit the viability and health of natural aquifers by increasing the burden on regional water demands and by creating significant impermeable surfaces as part of the infrastructure for algae cultivation.

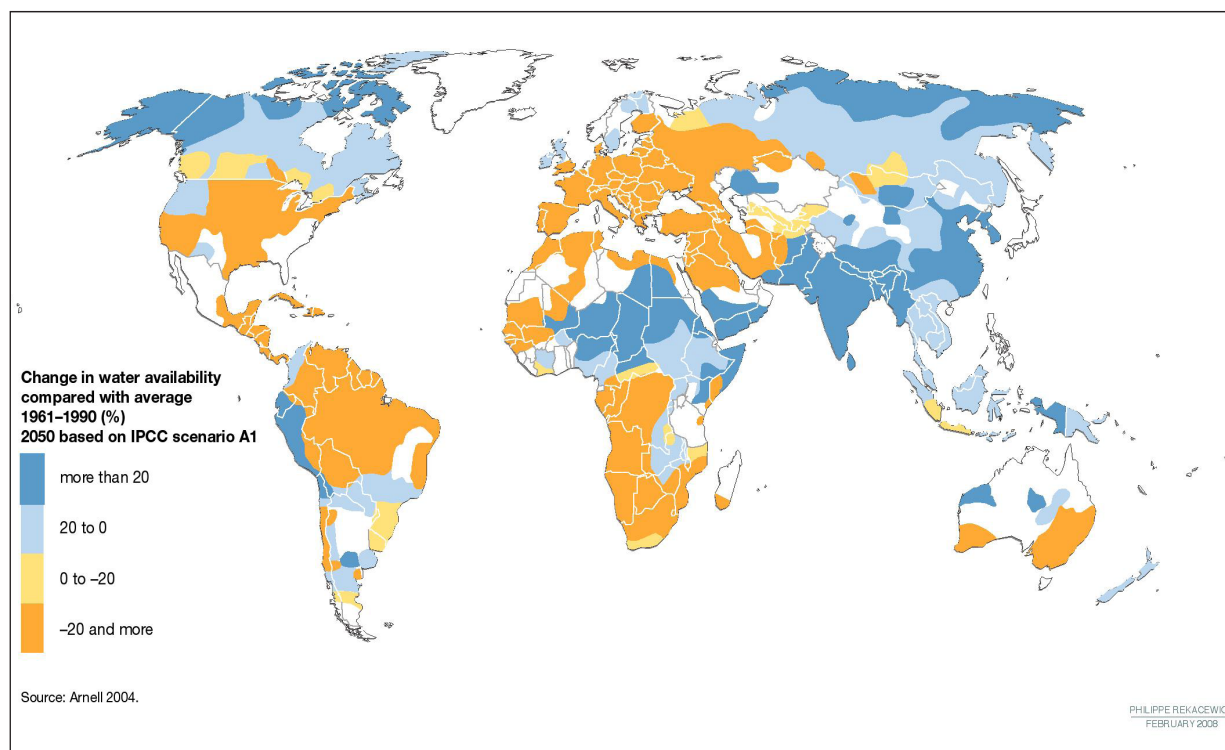
Infiltration of rainwater allows for aquifer recharge and is vital to the natural purification of water.<sup>8</sup> In this process, soil filters rainwater as it flows into the aquifer and also allows water to be retained on-site to nourish plant life and dependent herbivores. If consumption or redirection (runoff) of rainwater, such as by algae

cultivation facilities, exceeds recharge, the water table in the aquifer will most likely decline.

Historically, almost all agriculture and urbanization has reduced groundwater infiltration by increasing impervious surface area and interrupting the natural hydrologic cycle. The construction of a commercial-scale, (especially open pond) algae cultivation facility would decrease the biological activity in the soil directly below the site. However, the degree to which impervious surface impacts groundwater infiltration and the health of the greater watershed is also dependent upon the soil type and permeability, as well as water source and type.<sup>9</sup> The use of freshwater that is not permitted to filtrate naturally may greatly alter groundwater and aquifer levels, whereas the use of seawater may influence salinity of shallow freshwater aquifers, having a different yet serious impact on local or regional ecosystems.

Production facility discharge, runoff, and flooding near wetlands may put at risk adjacent swamps and vernal ponds that depend on a clean, cool, stable and reliable water source. Runoff especially high in chlorides from salinated water (causing groundwater salination), nitrates or phosphates (causing eutrophication), and discharge high in chemicals from harvesting processes threaten downstream water quality as well as that of the greater watershed.

Different pathways to production will have varying effects on water usage, downstream water, water quality, and groundwater and aquifer infiltration. Nevertheless, downstream water conditions, regardless of the pathway followed, will largely depend on the management of water during and after key production processes.



**Figure 1:** In the context of climate change and changing patterns of rainfall, the decrease of runoff water may put at risk large areas of arable land. The map indicates that some of the richest arable regions (Europe, United States, parts of Brazil, southern Africa) are threatened with a significant reduction of runoff water, resulting in a lack of water for rain-fed agriculture.

## Land

Given the potential to produce more gallons of biofuels per acre on nonarable land, algae cultivation promises a great advantage over biofuels derived from arable land crops. However, food security and land use efficiency are major components of potential land impacts and both could be improved or deteriorated by the externalities of the various algae-to-biofuel production processes.

### *Food Security*

Although there is debate over what constitutes “available” or “marginal” land acceptable for biofuel feedstock production, critics believe that designating arable farmland for energy production contributes to food supply constraints. The United States has been particularly culpable because one-fifth of the nation’s corn crops are currently being used for fuel production and about 20 million acres of farmland were diverted from soybean production to corn production for ethanol in 2007 alone.

Estimates suggest that algae grown on 1–3 percent of available crop area could theoretically meet 50 percent of the country’s transportation fuel needs, a significant contrast to the amount of land used by more conventional feedstocks.<sup>10</sup> More to the point, most algae cultivation does not directly require arable land.

### *Land Use Efficiency*

Land use efficiency will vary among different production processes. For instance, because of the defining characteristics of cultivation systems, open systems are likely to have a larger land use footprint than other photosynthetic, heterotrophic, or integrated biofixation systems for algae cultivation. The degree to which land use efficiency will influence the gallon per acre metrics of algae biofuel production is perhaps not yet calculable.

Maximum culture densities (grams of algae per gallon of culture) and proportion of oil in the final biomass will be essential to measuring land use efficiency and the scalability of algae cultivation systems. Liquid fuel products are typically measured in gallons per acre or liters per hectare (harvested) per year. Future analyses will need to differentiate between biomass yield, oil yield, and net energy yield of differing production pathways for two reasons: oil

yield per ton of biomass will vary from one algal species and production method to another, and algal oil is not the sole product to come out of algae biomass. Nevertheless, “per acre” estimates may serve as one of the defining parameters for land use in sustainable biofuels production.

## Soil and Biodiversity

Twenty percent of the total U.S. land area is used for crop-based agriculture and an even greater amount of land area (51.8 percent) accounts for all agricultural activities.<sup>11</sup> According to the U.S. Food and Agriculture Organization (FAO), there are “linkages between agricultural biodiversity and climate change.”<sup>12</sup> This places a large responsibility on farmers to protect soil quality and biodiversity. Although algae-based biofuel production can be conducted on underutilized, nonarable land, soil quality and the proliferation of flora and fauna in and around such (desert) ecosystems are still of vital importance.

### *Soil Quality*

Soil contamination results when hazardous substances are either spilled or buried directly in the soil or migrate to the soil from a spill that has occurred elsewhere. Soil can become contaminated from uncontained landfills, hazardous particulates released from smokestacks and deposited on the surrounding soil as they fall out of the air, and water that washes contamination from an area containing hazardous substances and deposits the contamination in the soil as it flows over or through it. Highly salinated water may also contribute to soil contamination in freshwater ecosystems (Figure 3).

### *Proliferation of Flora and Fauna*

Biodiversity, the number of different species in a given area, is important for survival of a habitat and is often used as a measurement of ecosystem health. In a balanced environment, weather, predators, diseases or parasites, and availability of food control population numbers of each species and ensure long-term balance. Land transformation, water and soil contamination, air pollution, and the introduction of invasive alien species threaten this biodiversity.

Chemicals and algae particles present in released waters from the cultivation and harvesting phases





ISTOCKPHOTO.COM/DESERTDIGITALIMAGES

**Figure 2:** Western Kingbird. Biodiversity and migratory patterns could be at risk when land transformation alters existing habitats.

of production could affect biodiversity at a regional level by threatening the stability of natural control mechanisms. Further, the salt left behind from algae cultivated in brackish water could be a potential source of soil and groundwater contamination. However, there is also the possibility for algae to improve soil and biodiversity where native species are cultivated and nonchemical methods are used for harvesting and other processes.



PHOTO CREDIT: KATARINA DORN

**Figure 3:** Soil contamination from saline water.

## Air

The various algae-to-biofuel production processes could affect air quality, a major component of regional and global environmental concerns. However, consensus on the scope of impact on air quality and evaporation rates by algae-to-biofuel production has not yet been reached.

### *Air Quality*

About four percent of deaths in the United States can be attributed to air pollution, according to the Environmental Science Engineering Program at the Harvard School of Public Health.<sup>13</sup> Air pollutants take the form of solid particles, liquid droplets, or gases and are typically associated with asthma, heart damage or failure, and cancer. The burning of fuels is a major source of air pollution. In addition to reducing dependency on fossil oil, reducing concentrations of pollutants is a primary objective guiding the development of biofuels.

### *Evaporation Rates*

The evaporation rate of water depends on water temperature, the amount of exposed surface, degree or rate of agitation, and the humidity of the air above the water. Large exposed surfaces of stirred water in a warm, dry climate will have greater evaporation rates.<sup>14</sup>

Whereas high concentrations of algae cells will likely decrease pond evaporation rates, as higher concentrations decrease the amount of water escaping from below the pond surface to the air above, evaporation from vast commercial-scale open pond cultivation systems could potentially affect local and regional humidity and alter precipitation patterns, native habitats, and local ecosystems.

## Energy and Carbon Balance

Given the lack of a leading, industry-recognized production pathway, the potential energy balance of algae biofuels is a highly uncertain calculation, and heavily dependent on a reliable assessment of energy flows, inputs, and yields for each fuel production system. Similarly, estimates for the balance of carbon emissions associated with algae biofuel production range widely based on the algal species grown, the types of biofuel produced, the amount of and type of fossil fuel displaced, and the energy savings realized by the

production of coproducts or biological services such as wastewater treatment.<sup>15</sup> Energy balance considerations are closely related to carbon calculations, but they are not synonymous and must not be conflated.

### *Energy Balance*

The energy balance in algae biofuels can be quantified by comparing energy inputs and losses: energy consumed or released to produce a particular biofuel compared with the embodied energy of that biofuel (as a market-ready product).<sup>16</sup> With respect to algae biofuel production, energy inputs may include electricity, heat, pressure and other energy needed to operate technologies such as water lifting, paddle wheels, illumination (artificial light), harvesting mechanisms, biomass transfer to conversion facilities, gas transfer systems, reactors, and systems maintenance. A more thorough assessment of energy inputs will include not just those needed for the production of the fuel but also the energy consumed in the pretreatment of feedstocks, chemicals and other catalysts, environmental remediation because of contamination by processing methods (e.g., chemical recovery and wastewater treatment), and even the fabrication of materials and technologies used by the production process, such as pond liners or photobioreactors, light bulbs, and filters.

Many aspects of algae-to-biofuel production can influence the energy balance and each stage in the production process could have far-reaching impacts on the rest of the production chain.<sup>17</sup> For example, very high energy yield calculations usually come from assuming that all algal growth is in the form of high-energy components such as TAG lipids, yet this is not always the case.<sup>18</sup> The algal species (e.g., ratio of carbohydrates to TAG lipids), culture conditions, and downstream processing methods utilized will also influence the energy yield of an algal biomass fuel product.

Energy balances can either look at total energy including solar energy used in the production of algae biofuels or be limited to the fossil fuel energy used. Total energy is ultimately more important and is vital when comparing different algae production processes. However when comparing algae biofuels to other energy systems, it is important to decide what aspects of energy are most relevant. If one is using energy balance as a metric of fossil fuel avoidance,

fossil fuel inputs are sufficient. Such a calculation is a better proxy for carbon balance but still an imperfect alternative.

### ***Carbon Balance***

In accounting for carbon balance—or better, the greenhouse gas balance—of algae, it is critical to consider the net impact of the specific biofuel production pathway on atmospheric concentration of heat trapping gasses. As with the production of petroleum fuel or first-generation biofuels, carbon dioxide (CO<sub>2</sub>) and methane (CH<sub>4</sub>)—two of the earth’s most abundant GHGs—are byproducts of various algae-to-biofuel conversion processes.<sup>19</sup> However, as with the measurement of energy inputs to calculate energy balance, the level of GHG abatement achieved by a production pathway will largely depend on the circumstances—particularly on the CO<sub>2</sub> released from individual production processes employed—and thus cannot be generalized. The capacity of algae to measurably mitigate industrial GHG emissions and contribute to the carbon balance promises to be a closely studied calculation, with significant emphasis on the overall quantity of GHGs captured versus those released.

The primary carbon and energy benefits from algae biofuels involve two separate displacements—direct and process-related. Fuel derived from algae displaces fossil fuel (i.e., leaves the fossil fuel in the ground). What is important in determining the overall carbon balance from this displacement is calculating all of the carbon (direct and indirect) involved in the lifecycle of algae production, including end use. If these emissions are fewer compared to the overall lifecycle carbon balance of extracting and burning fossil fuel, then it is appropriate to discuss the negative carbon balance of algae biofuels relative to petroleum.

In this calculation, it is important to note that one source of emissions that algae cultivation is expected to avoid are emissions from land-use change. Converting carbon rich forests or grasslands to agriculture usually results in the release of most of the carbon stored in these landscapes. Crop-based biofuels can cause this conversion directly if these crops are planted directly on converted land or indirectly by competing for arable land and causing other crops to move to converted land. Algae, in contrast, does not benefit from being cultivated on arable land and is not expected to compete for such land.

The second displacement, which is process-related, involves the displacement of carbon emissions by the co-products or byproducts of the algae biofuels process. For example, growing algae in a process that also treats wastewater displaces the carbon that would have been generated in conventional wastewater treatment processes. In doing so, as van Harmelen and Oong (2006) explain,

“Approximately one ton of algae biomass produced during wastewater treatment reduces the equivalent of one ton fossil CO<sub>2</sub> derived from the algal biomass and the GHG reductions compared to conventional wastewater treatment processes, as well as fertilizers and other potential coproducts, currently derived from fossil fuels.”<sup>20</sup>

### ***Use of Carbon from Carbon-Emitting Facilities***

The capacity of algae to absorb and convert CO<sub>2</sub> emissions from fossil fuel combustion into a carbon-based biofuel has been investigated for its potential role in GHG reduction efforts. All biomass-to-energy processes involve some amount of atmospheric carbon cycling. Algae, however, absorb or “fix” CO<sub>2</sub>—photosynthesis allows algae to store energy from sunlight (in the form of sugars and starches)—with greater efficiency than such higher plants as trees and agricultural crops.<sup>21</sup> Some algal strains have the capacity to fix high levels of carbon CO<sub>2</sub>,<sup>22</sup> producing 1 unit (ton) of algal biomass is estimated to fix 1.5–1.8 units of CO<sub>2</sub>.<sup>23</sup>

Although CO<sub>2</sub> is a necessary input of algae growth, some cultivation systems rely on atmospheric CO<sub>2</sub> and others require artificial CO<sub>2</sub> inputs. Where artificial inputs are necessary, CO<sub>2</sub> can be provided by power plant and industry flue gases, and on-site use of biogas derived from wastes. This biogas can be continuously recycled in the biofuel production process. For example, waste CO<sub>2</sub> from fermentation or combustion processes can be captured and used in the cultivation stage to boost growth productivity of new biomass. In other words, commercial-scale carbon fixation may be a potential route to reducing GHG emissions by way of capturing industrial CO<sub>2</sub> emissions through algae cultivation systems and then using the algae to displace fossil fuel inputs to the industrial process.<sup>24</sup>

However, it is critical to understand that from the perspective of net impact on atmospheric GHG concentrations, it does not actually matter if CO<sub>2</sub> is first released to the atmosphere from an industrial

facility and then captured by algae or captured directly from the facility flue gas by the algae. In fact if the energy needed to utilize flue gas directly into the algae cultivation system does not produce sufficiently more incremental algae than what would have occurred using atmospheric carbon, then the carbon balance will be better if the two systems are not coupled.

The GHG benefit in this process comes from not using fossil fuels and this benefit must only be counted once. In other words, either the algae gets the credit for displacing the fossil fuels or the industrial facility gets the credit, but there can only be one credit. If an algae facility uses carbon from an emissions source that is covered by climate regulations (e.g., coal powerplant, cement plant, etc.), then either the covered entity can claim a carbon credit, in which case the algae fuel company must treat all carbon emissions associated with its process (including combustion of the algae) as new pollution in the atmosphere, or the algae fuel company can claim the credit, while the covered entity obtains carbon allowances for its carbon emissions. What cannot happen is that both the covered entity and algae biofuel facility each claim credit for offsetting carbon—this would be considered “double counting.”

Terminology is also important in discussing both energy and carbon balance. Terms such as “recycling”<sup>25</sup> are not accurate as recycling suggests that fossil carbon that would have otherwise ended up in the atmosphere will not. Again, the benefit lies in displacing fossil fuels with algae biofuels, not from using fossil-derived carbon instead of atmospheric carbon as a feedstock.

“Sequestration” is also a term used frequently within the algae industry and media.<sup>26</sup> Carbon dioxide released from the burning of coal, oil, and natural gas is carbon that, once released into the atmosphere, adds to the current carbon account. Since algae simply cycle the carbon, growing algae with CO<sub>2</sub> captured from power plants is not true “sequestration” unless the algae is then trapped where the CO<sub>2</sub> cannot escape (e.g. buried). Otherwise, the algal biomass that is converted into fuel will eventually be burned, releasing CO<sub>2</sub> into the atmosphere.

The total industrial utilization of CO<sub>2</sub> in the United States, as of June 2001, was about two percent of the CO<sub>2</sub> generated from power plants (Table 1).<sup>27</sup> The utilization of CO<sub>2</sub> from flue gases is relatively well understood;<sup>28</sup> however, its application in conjunction with GHG abatement and algae cultivation may still

<b>Table 1. Summary of Availability of CO<sub>2</sub> Sources</b>	
<b>CO<sub>2</sub> Source<sup>32</sup></b>	<b>Potential CO<sub>2</sub> (109 kg/y)</b>
Concentrated, high-pressure sources:	
Liquid synthetic fuel plants	40
Gaseous synthetic fuel plants	220
Gasification/combined-cycle power plants <sup>33</sup>	0–790
Concentrated, low-pressure sources:	
Enhanced oil recovery	8–32
Ammonia plants	9
Ethanol plants	<0.1
Dilute high-pressure sources:	
Noncommercial natural gas	52–100
Refineries	13
Dilute low-pressure sources:	
Anaerobic digestion (biomass/wastes)	230
Cement plants	26
Fossil steam plants	0–790
<b>TOTALS:</b>	<b>600–2,250</b>



be in need of research and development.<sup>29</sup> It is fairly uncommon today to find cultivation facilities coupled with power plants.<sup>30</sup> This is largely due to the nascence of the industry, but may also be due to a number of variables such as land availability in relative proximity of power plants, the volume (percent) of industrial CO<sub>2</sub> an algae system can realistically utilize, the need for emissions cooling (flue gases are too hot for algae), the risk of impurities such as concentrations of heavy metals in industrial CO<sub>2</sub> emissions, and other complexities associated with CO<sub>2</sub> capture.<sup>31</sup>

### ***Co-location***

Algae biofuel production could potentially further improve its energy and carbon balance by co-locating its production facilities. Individual process energy balances for algal cultivation, harvesting, and oil extraction processes may be improved by co-locating with each other, with power plants, and with regional terminal racks. Co-location of these processes could minimize heat losses and reduce emissions caused by the transportation of intermediary products. It is still unclear, however, as to what extent co-location could affect the overall energy balance of algal biofuel production.

## CHAPTER 2

# Mapping Pathways for Algae-to-Biofuel Production

Algae-to-biofuel production represents a complex intersection of industries. Therefore, the mapping of production pathways must be simplified in order to identify the big picture environmental benefits, concerns, and unknowns in each commercial-scale production process. The pathway approach maps existing and potential pathways for algal biofuel production and, where applicable, pathway variations. This approach was selected for the visual clarity in which environmental externalities could be conceptualized from a nontechnical perspective and without prematurely devaluing or championing any specific process in the nascent algal biofuel industry.

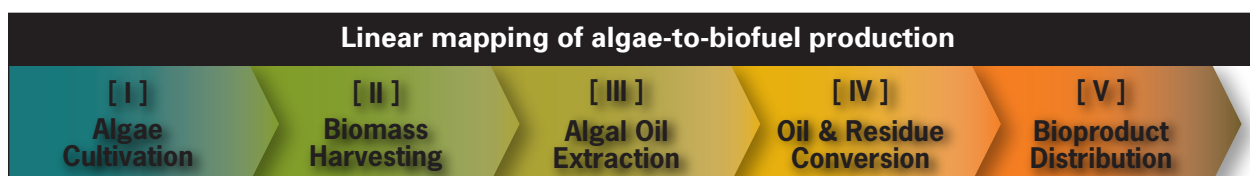
## Algae-to-Biofuel Production Pathways

### *Description of Pathway Framework*

Algae-to-biofuel production is divided into four stages, including algae cultivation, biomass harvesting, algal oil extraction, and oil and residue conversion. Because of the limitations of the scope of this report, the pathway framework does not represent the full life cycle ("field-to-wheels" or "well-to-wheels") of an algae biofuel product; the origin and destination, including bioproduct distribution (the fifth stage), of some

inputs, outputs, and products will not be discussed at great length.

Each of the first four stages is further broken down into basic, individual, or multiple processes to explain the primary components of algal biofuel production that may have positive or negative environmental externalities (Figure 4). Identified environmental benefits, concerns, and unknowns are noted at the base of each pathway process. By examining the inputs and outputs of a particular pathway, or process within



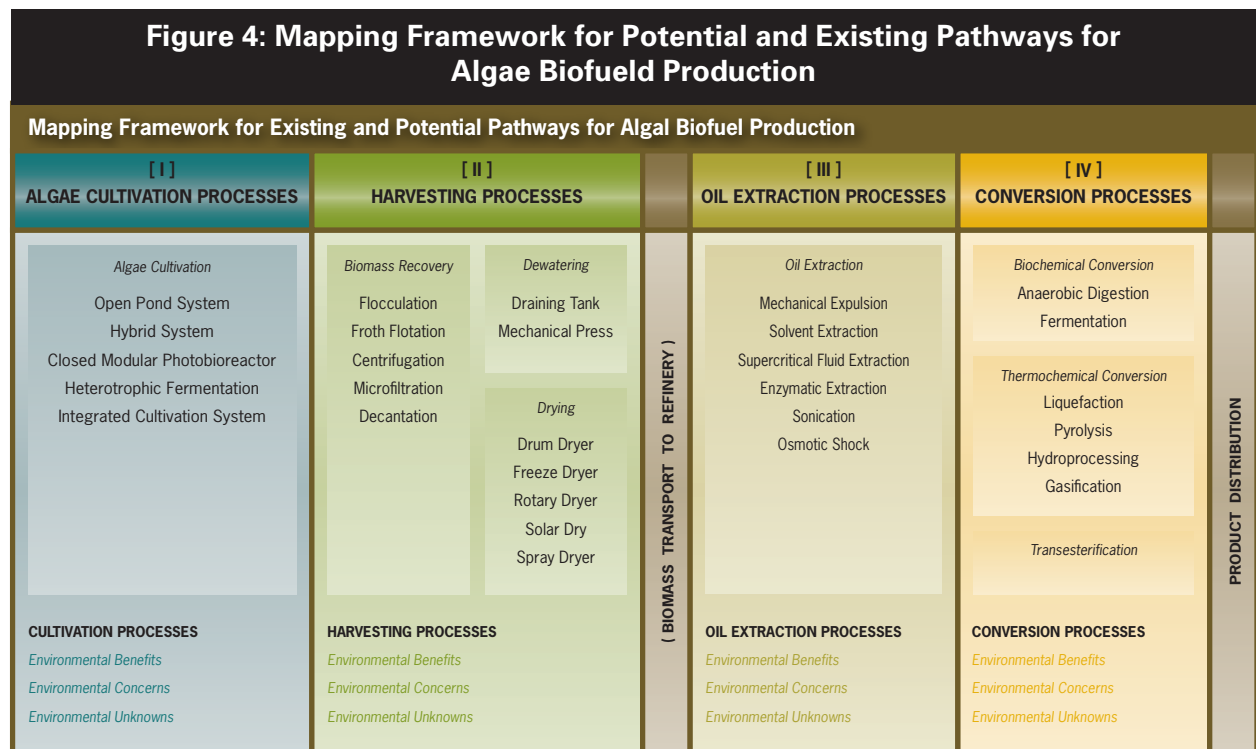
a pathway, these environmental externalities can be anticipated and appropriately addressed.

The flexibility in mapping production pathways allows for easy manipulation or addition of alternative pathways, processes, and inputs. Eventually, feedstock pretreatments, system fabrication and materials, system operation and maintenance, and other indirect inputs should be analyzed to accurately reflect all direct and indirect resource inputs and outputs, the true energy balance of algae-to-biofuel production and, ultimately, the complete life cycle impact.

For the purpose of this report and mapping exercise, “coproducts” refers to process outputs (e.g., biofuel, animal feed) that have an existing or emerging application (market). “Byproducts” refers to process outputs with little or no value (in the existing market), that must be disposed of (e.g., CO<sub>2</sub>, tar, certain acids), or for which pretreatment is required in order to reclaim value or viable reuse (e.g., wastewater).

Not all pathways will adhere to this structural framework. Some pathways may employ more than one process at the same time or may skip an entire process altogether. Pathway processes are often interchangeable and, therefore, those mapped here in no way represent the full range of existing and potential processes, nor does it showcase every technology and technique discussed in this report. Additionally, some processes may appear in more than one pathway. This is especially true of biomass harvesting and oil and residue conversion processes. In such a case, environmental externalities are the same in each instance.

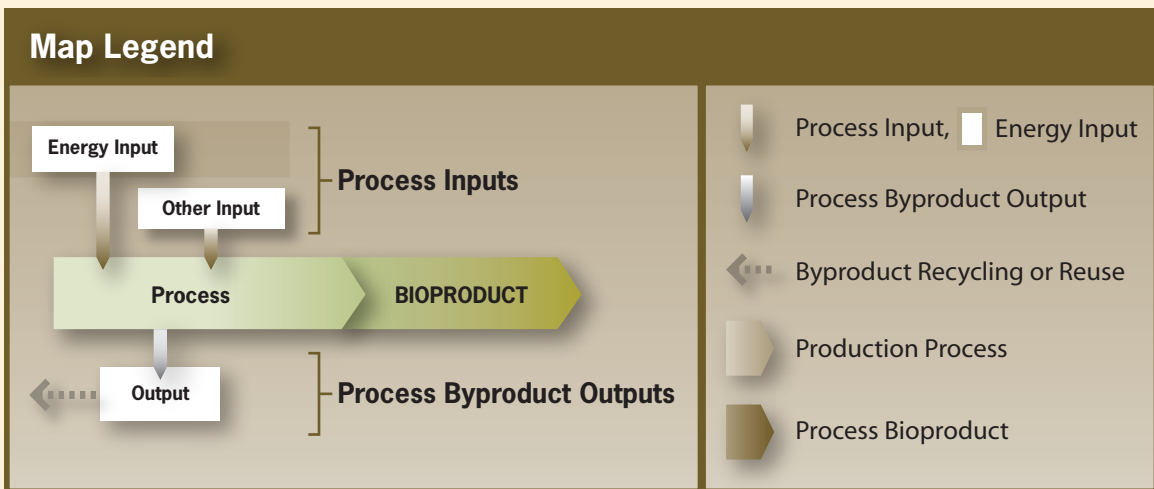
Finally, because of the breadth and variety of existing and potential production pathways, this report does not attempt to give all available information on each known system. This would be an exhaustive and perhaps naïve effort to present systems that may be constantly evolving or soon be obsolete or abandoned.



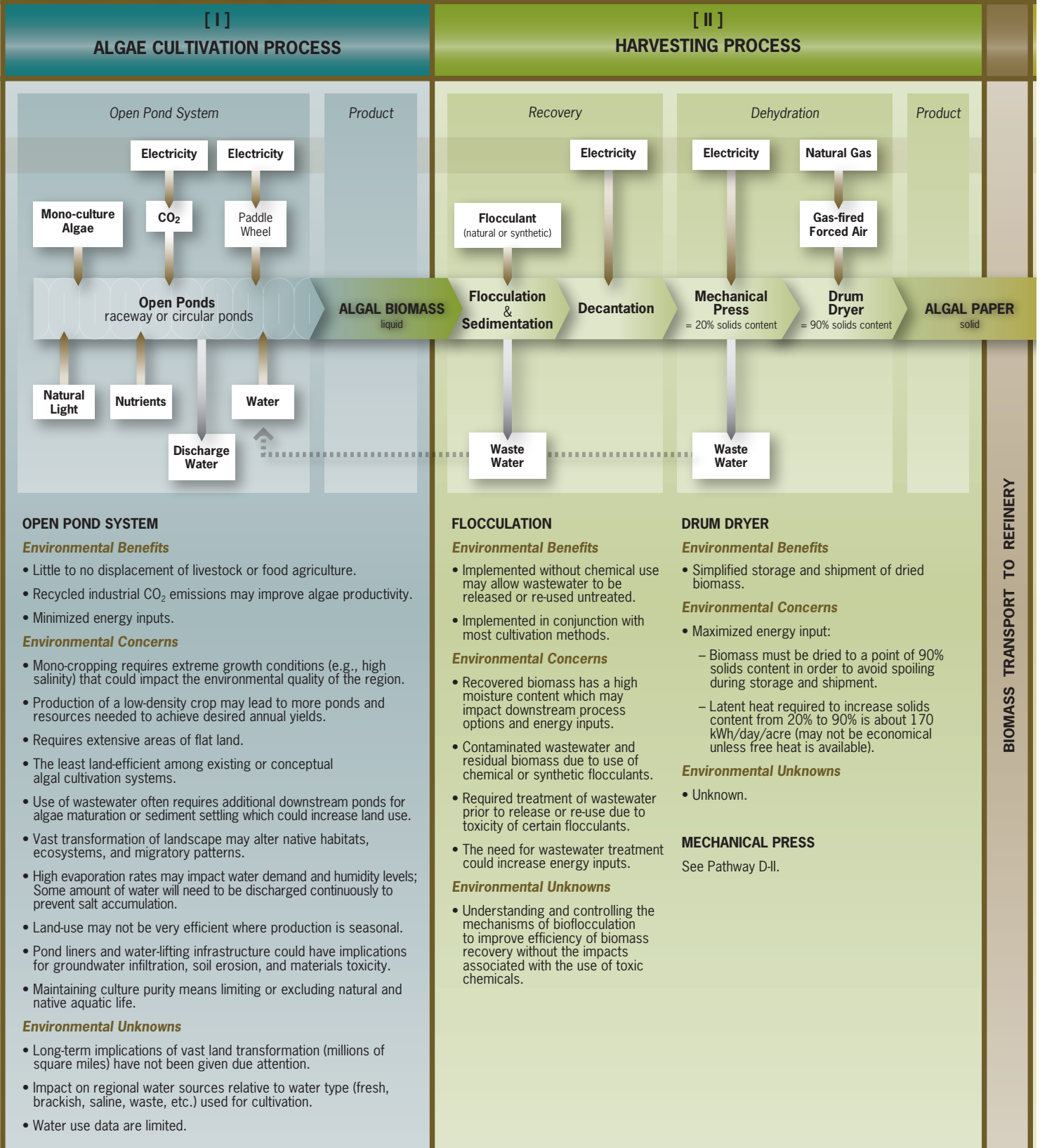
### Algae-to-Biofuel Production Pathway Maps A–E

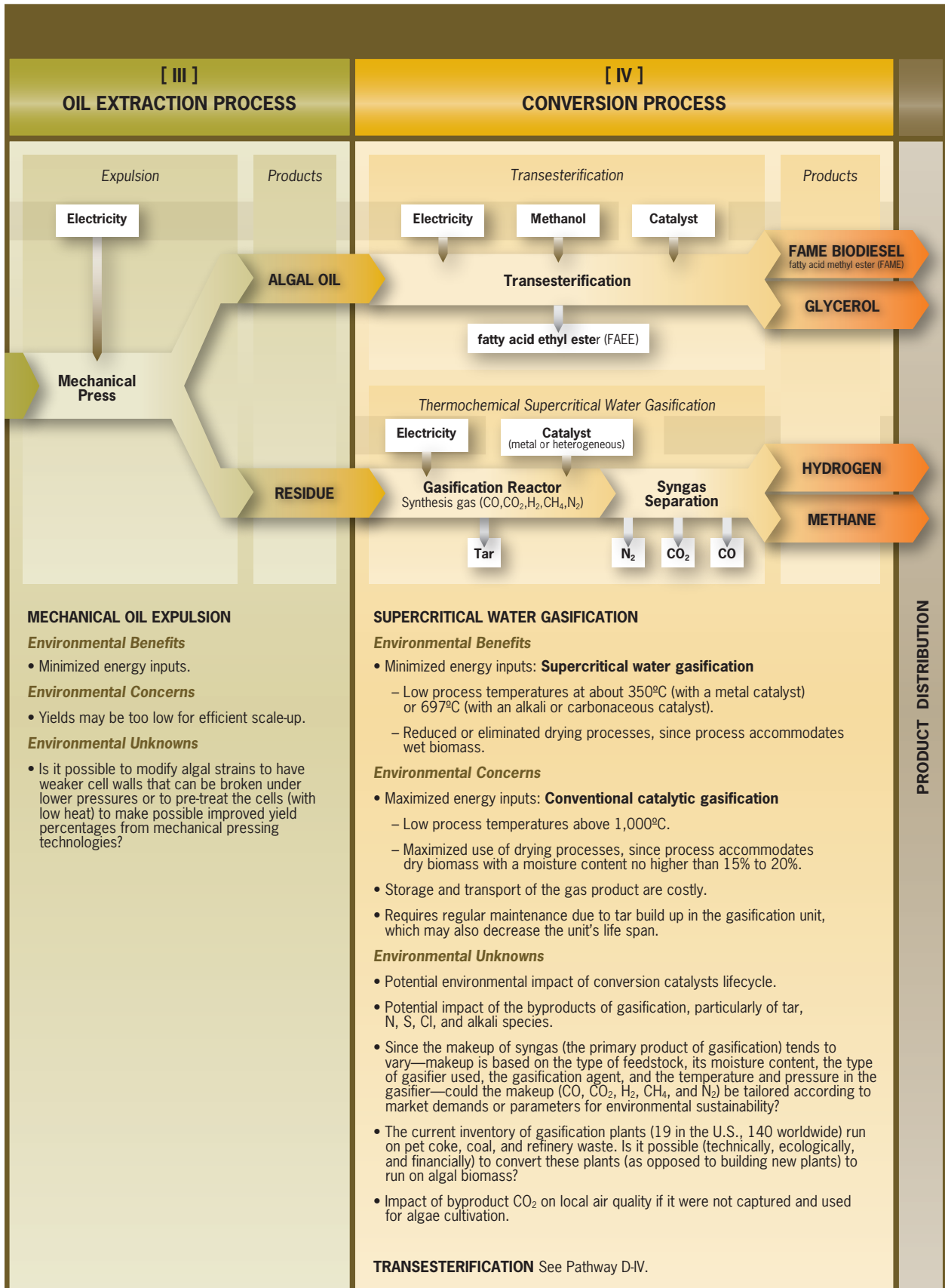
Five existing or potential pathways (Pathways A–E) to algae-based biofuel production are mapped out in the following composition of processes:

- A. OPEN POND SYSTEM  
→ Flocculation → Mechanical Press → Drum Dryer → Mechanical Oil Expulsion → Gasification → Methane
- B. HYBRID SYSTEM  
→ Microfiltration → Mechanical Press → Liquefaction or Fast Pyrolysis → Hydroprocessing → Green Diesel
- C. MODULAR (INDOOR) CLOSED PHOTOBIOREACTOR  
→ Sonication → Fermentation → Methanol
- D. HETEROTROPHIC FERMENTATION  
→ Centrifugation → Mechanical Press → Rotary Dryer → Solvent Extraction → Transesterification → Biodiesel
- E. INTEGRATED CULTIVATION SYSTEM  
→ Solar Drying → Supercritical Fluid Extraction → Modified Fermentation → Butanol + Hydrogen

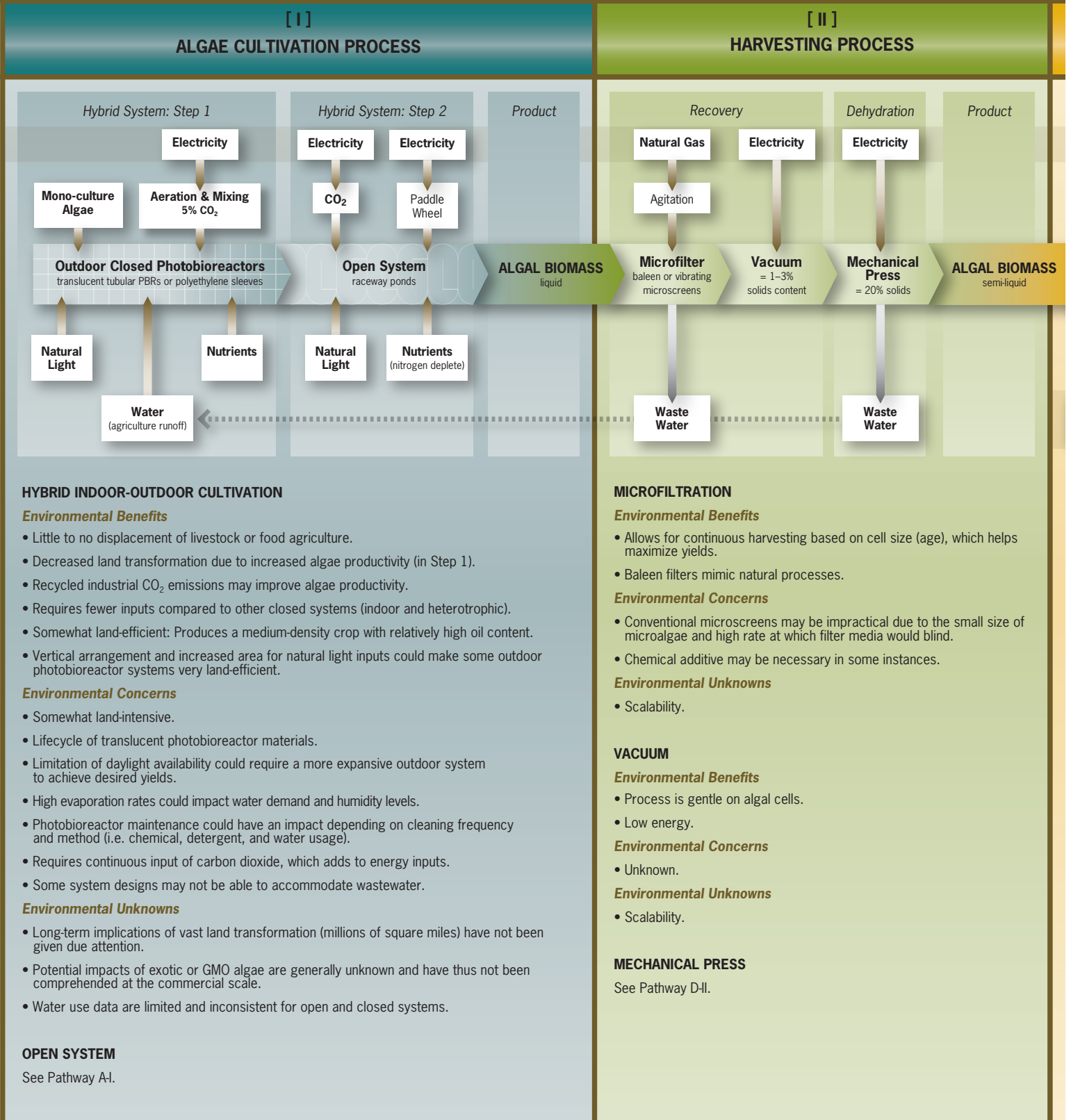


**PATHWAY A: Open Pond System**

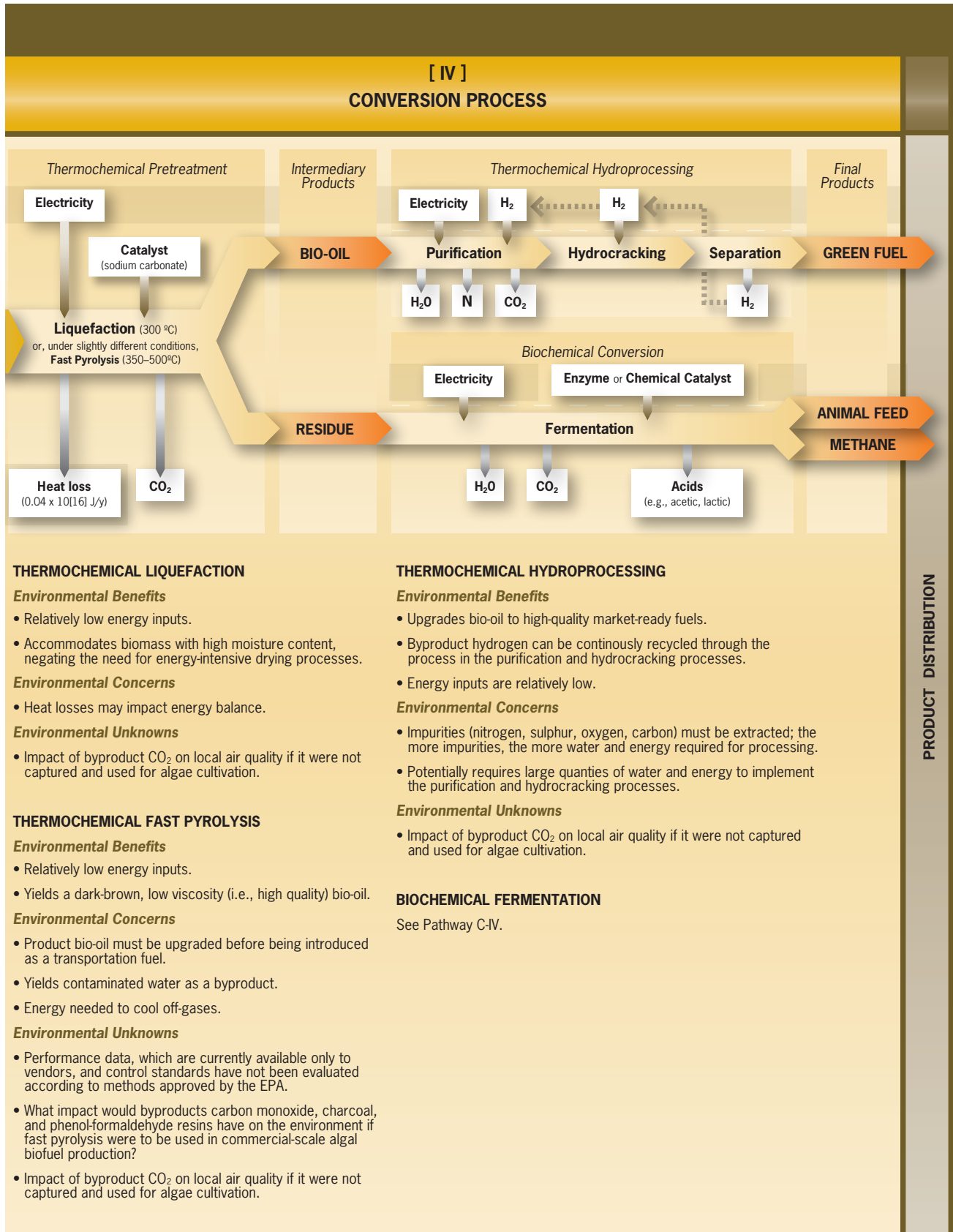


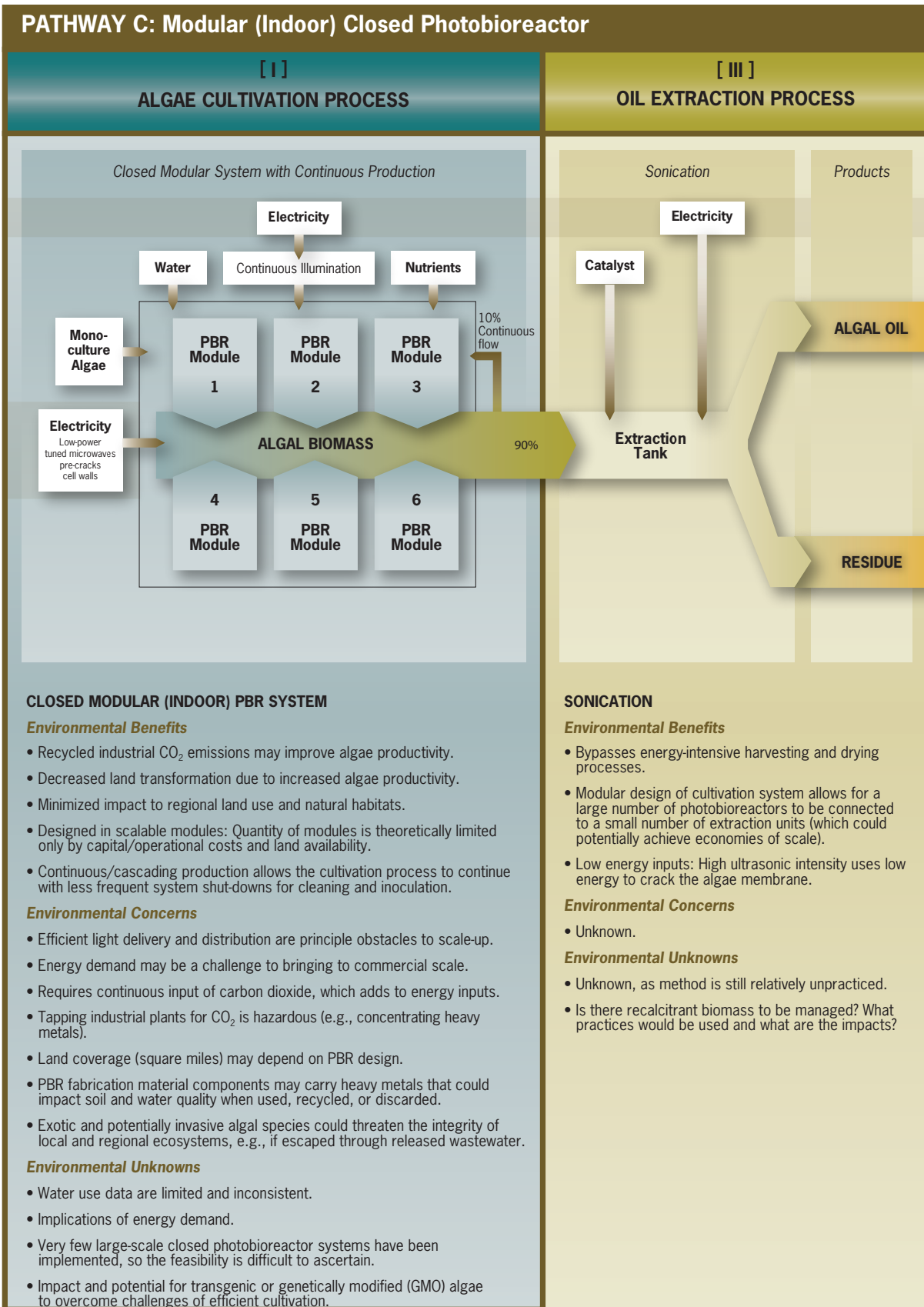


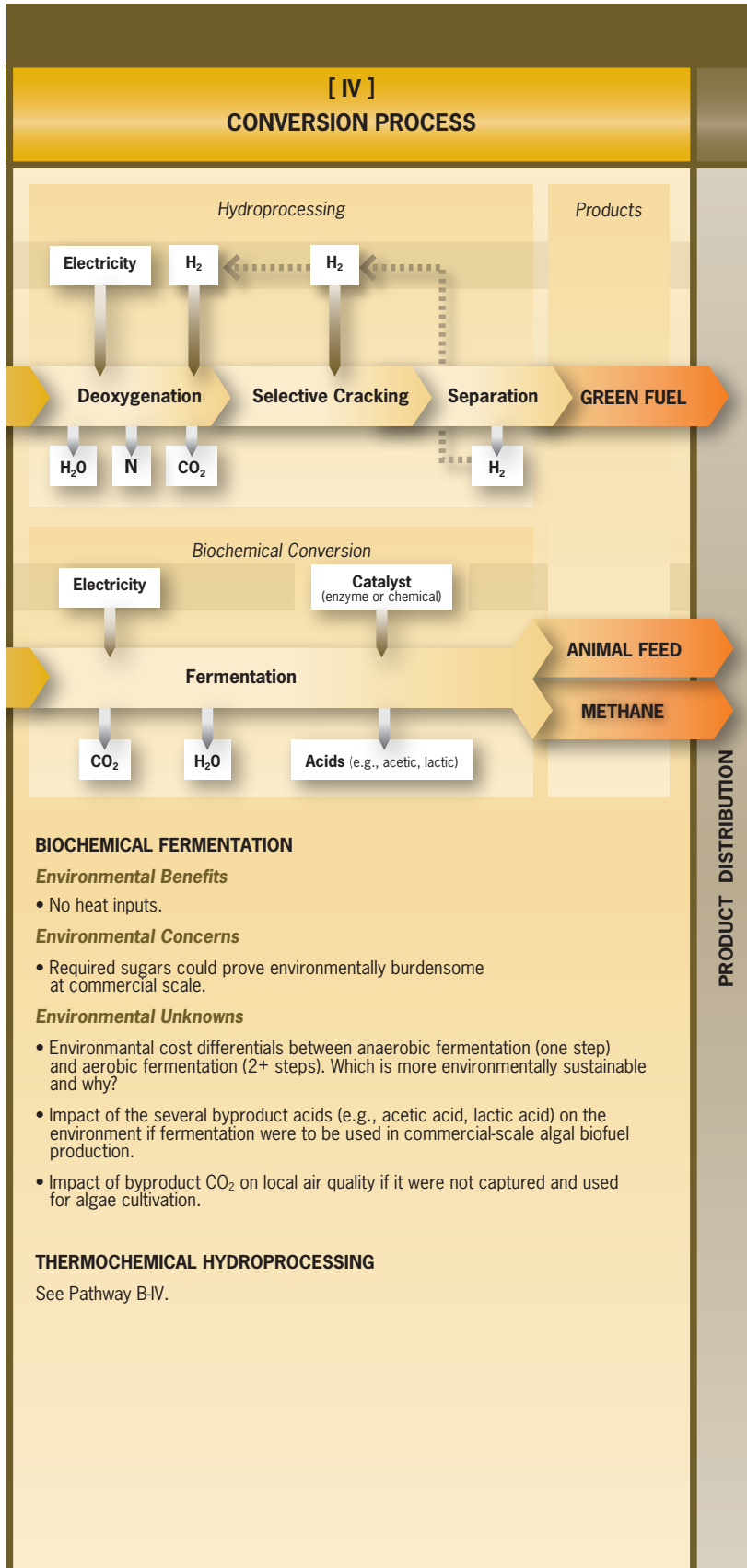
**PATHWAY B: Hybrid System**



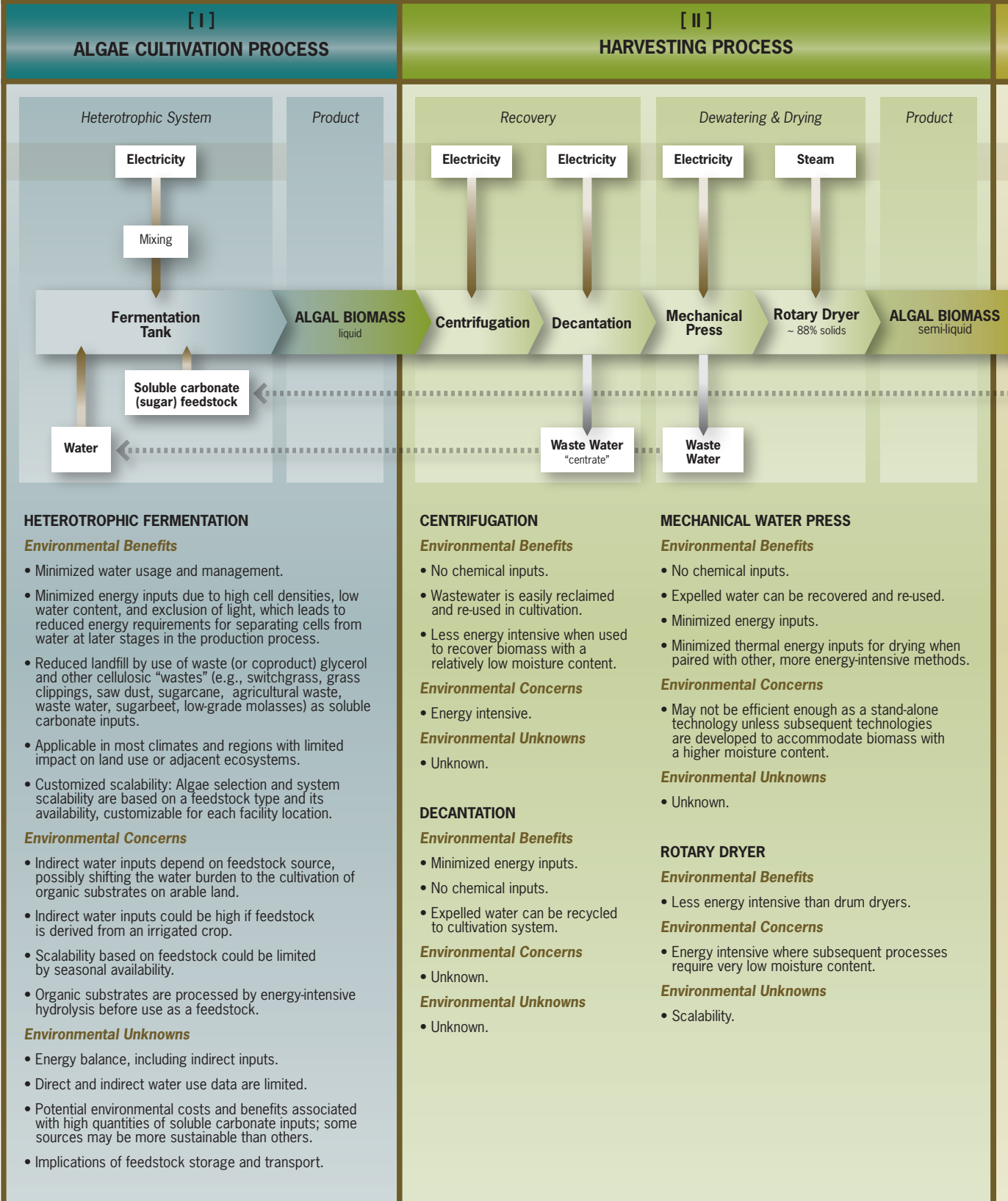


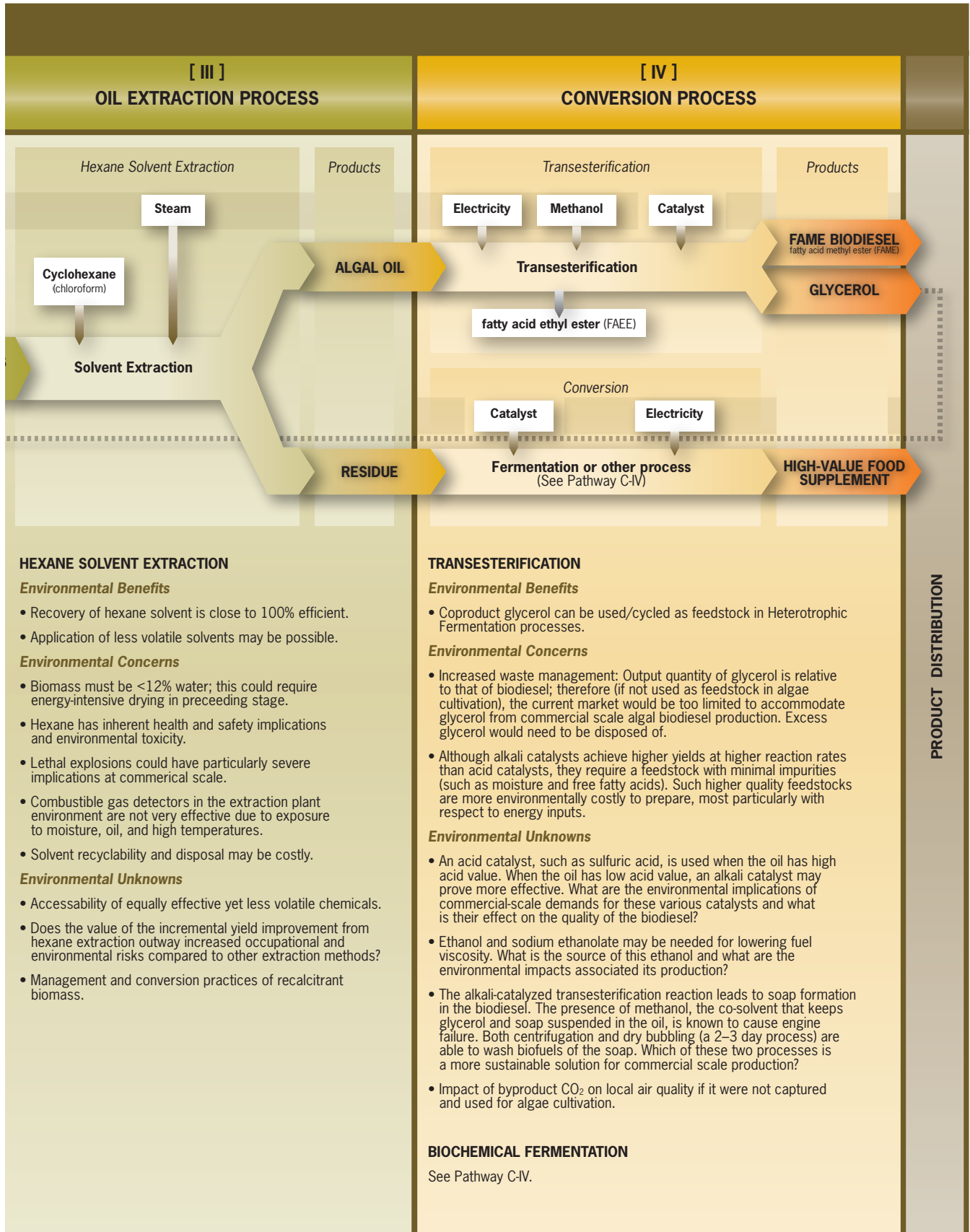


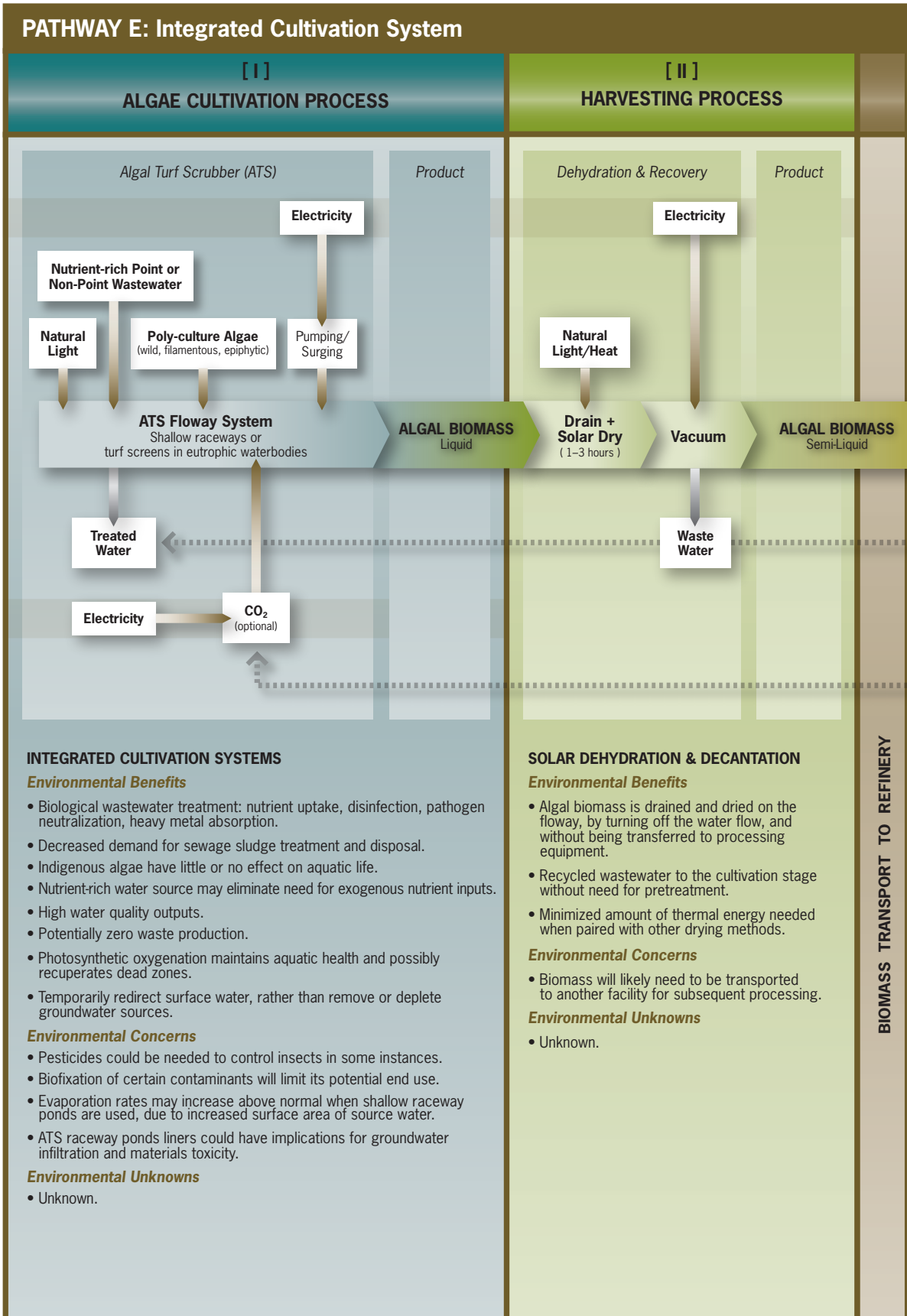


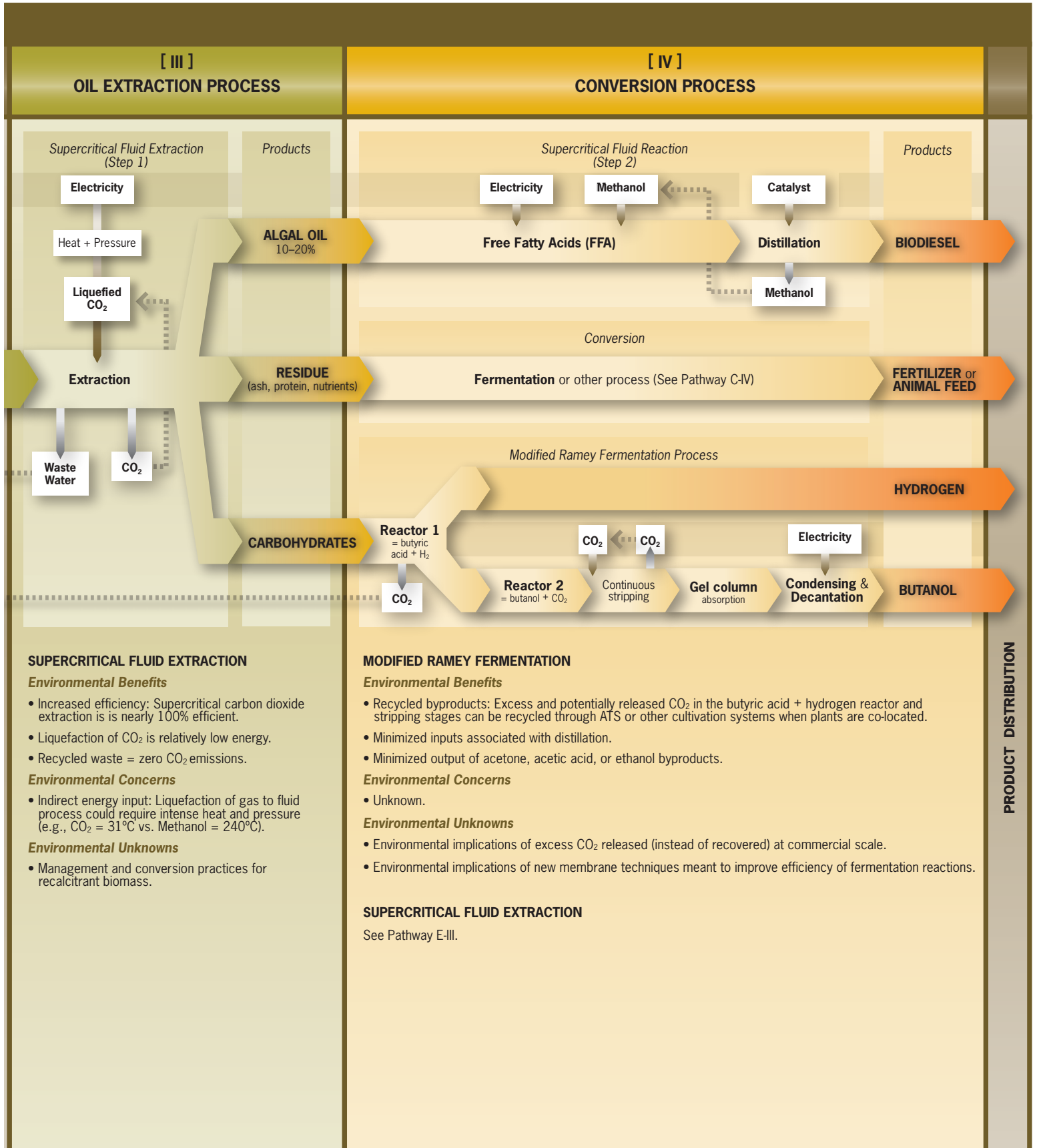


**PATHWAY D: Heterotrophic Fermentation**











## CHAPTER 3

# Exploring the Stages of Algae-to-Biofuel Production

In this report, we explore each of the primary stages of algae-to-biofuel production and the associated environmental implications. Based on available peer-reviewed journal articles and other scientific publications, news releases, industry workshops, and personal communications with academic experts and entrepreneurs, we have summarized the state of knowledge around the environmental challenges of individual production pathways. The primary stages are categorized as: 1) pathways for algae cultivation; 2) pathways for biomass harvesting; 3) pathways for algal oil extraction; and 4) pathways for oil and residue conversion.

## Pathways for Algae Cultivation

Algae are an attractive biofuels feedstock compared to other biofuel sources. Their rapid growth rate (doubling in 6–12 hours), high oil content (4–50 percent or greater of nonpolar lipids), biomass harvest (100 percent), and nonseasonal harvest intervals have led to claims of algae biofuel yields that are theoretically orders of magnitude higher than other biofuels feedstock.<sup>34</sup> Nevertheless, the diversity of algal characteristics and lack of scientific and industry consensus have so far made it difficult to forecast the true potential of algae as a fuel feedstock.<sup>35,36</sup>

The purpose of algae cultivation is to grow raw algal biomass for the downstream production of fuel, based on the oil and residual components found in the biomass. In order to flourish, algae need water, carbon dioxide, and essential nutrients, which are collectively referred to as the culture medium; algae cultivation facilities need land or other area to occupy; and, in

most cases, algae need light to drive photosynthesis. The varying manners in which water, nutrients, land, and light are supplied and managed for cultivation will have some effect on the environment, especially at the commercial scale.

One of the first steps in understanding the potential environmental impact of algae as a mass-produced biofuels feedstock begins with the cultivation process, where algae are grown by a variety of methods. Historically, the two primary classifications for algal cultivation systems are open systems and closed photobioreactors. Closed (photosynthetic) cultivation systems can be further subdivided into indoor and outdoor photobioreactors. Variations on cultivation systems have also emerged, such as hybrid (combined open and closed) cultivation, heterotrophic cultivation (without light), and integrated biofixation systems. In addition, there are other cultivation systems being implemented and new technologies being explored,

The balance of information provided for each of the various production stages is based upon the following premises:

- ▶ There is a preponderance of recent information available on algae cultivation practices.
- ▶ There is a general understanding that the cultivation of algal biomass could be one of the most resource-intensive stages in the production of algae-based biofuels, with a potentially significant environmental impact, particularly with respect to water, land, soil, biodiversity, air, and energy.
- ▶ The latter processes in a production pathway (e.g., oil extraction, oil and residue conversion) are not unique to algae. In addition, scientific literature and industry awareness concerning the environmental impacts of these technologies is steadily growing; however, emerging technologies for algae cultivation and biomass harvesting (and, to a lesser extent, oil extraction) are relatively unique with perhaps the greatest potential for developing in a sustainable manner.

including offshore cultivation, aquaculture, and ethanol sweating, which could become viable pathways as the industry develops; however, they are not within the scope of this report.

These five pathways—open systems, hybrid systems, closed photobioreactors, heterotrophic fermentation, and integrated biofixation systems—have been selected for discussion because they best represent the myriad approaches currently being researched and implemented for algal biomass production. Although thorough techno-economic comparisons have not been made among these five pathways and there remain considerable unknowns as to the economics behind each one, this section will provide an overview of the pathways, including system characteristics and core environmental issues as they relate to a scalable biofuels industry.

### OPEN SYSTEMS

Open systems, often implemented for their technical simplicity and relative affordability, are the most common method of cultivation today.



**Figure 5:** Open raceway ponds with paddle wheels (far right) for circulating the water.

Source: Seambiotic, Ltd.

### System Characteristics

Open systems are comprised of one or several shallow ponds—preexisting or man-made—that are exposed to the atmosphere, either outdoors or sheltered in greenhouses. They can take a variety of forms such as circular, lagoon, or raceway, the latter of which is the most common open system used for commercial algae cultivation.

Modern, commercial-scale open systems are typically designed as high- (growth) rate algal ponds (HRAP) in raceway formation (Figure 5) with a paddle wheel, wave pump, or baffles for circulating water with nutrients, gases, and algae (see Pathway Map A-I). Circular ponds, extensive ponds, and aerated lagoons are also commonly implemented, though not necessarily with the same mixing capabilities. Open pond systems operate in several locations throughout the world. In the United States, there are many examples of established pond systems, such as Earthrise Farms (California), HR Biopetroleum and Cyanotech Corp. (Hawaii), and Green Star Products, Inc. (Montana).

Optimal design parameters for large-scale, open pond cultivation have been known for many years.<sup>37</sup> The primary inputs to open pond systems are algae, light, nutrients, and water.

### Industry or Agriculture?

Algae cultivation is often characterized as an agricultural process and, in most cases, this holds true for the cultivation of algae for biofuel production. As with land crops, temperature, climate, sunlight, and other factors determine crop quantity (e.g., ears of corn per acre) and quality (e.g., vintage of wine from vineyard grapes), as well as the degree of associated environmental impacts, such as from fertilizer and pesticide usage, erosion and topsoil loss, and changes in water quality.

Therefore, the consistency and control of a cultivation process and output are critical in determining whether a process is agricultural or industrial. Though modern agriculture provides relative consistency, yields tend to vary by season and region. This will be even more so for algae, where cultivation and harvesting occur year-round in different climate conditions. For example, variations in light and temperature affect photosynthetic processes that in turn impact system outputs. Photosynthetic algae cultivation systems (e.g., open ponds, integrated wastewater/cultivation systems, and closed photobioreactors using natural light inputs) dependent upon these natural conditions are more likely to be categorized as agricultural processes. During heterotrophic cultivation and perhaps some types of photosynthetic cultivation (e.g., closed photobioreactors using artificial illumination or GMOs), inputs and climate conditions can be controlled in a manner that provides a very consistent process and product. Since external climate conditions and the availability of natural light do not impact the output product, these processes are more likely to be categorized as a type of industrial agriculture.

#### Algae

Open pond systems are typically designed for photoautotrophic monocultures.<sup>38</sup> At a given temperature, most algal species share similar environmental parameters, including abundant light, ample nutrients, and a pH that is characteristic of the growth medium.<sup>39</sup> Most algal species cultivated commercially in open systems (i.e., *Chlorella*, *Spirulina*, and *Dunaliella*),

while not necessarily for the purpose of biofuels production, are grown in highly selective, open-air environments that remain relatively free of contamination by other algae and protozoa.<sup>40</sup>

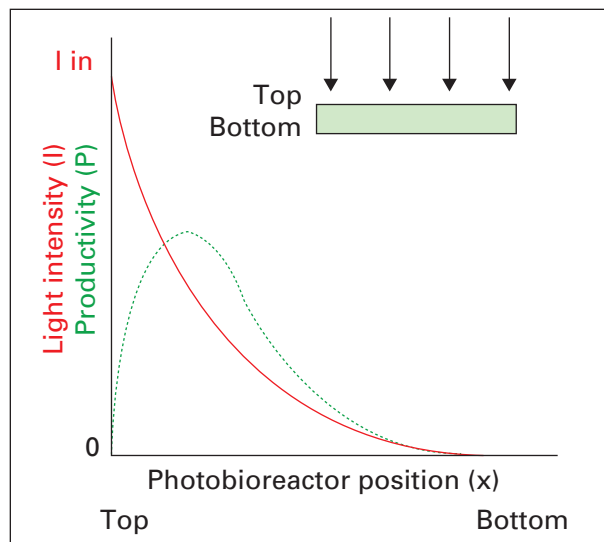
One of the main disadvantages of open systems is that parameters are harder to control than in closed systems. Management of environmental factors is very important in maintaining pure monocultures in open ponds. Because of a long light path, relatively poor mixing, and low photosynthetic efficiency, which lead to low biomass concentration and volumetric productivity, the algae growing season is largely dependent on location and is limited to warmer months when more light is available. Nevertheless, open ponds are the most common, commercially used algae cultivation systems in operation today.

Since the system is open, the culture is vulnerable to contamination. Contamination in open ponds is often described as predation or predominance of unwanted algal species or strains, algal weeds, microbes, or other nonalgal organisms. Bacterial and viral diseases (phycodnaviruses) could potentially pose an even greater threat to the integrity of large-scale monocultures.<sup>41</sup> Algal culture exposed to contamination can result in decreased quality and yield of the biomass. A sterile environment with controlled parameters, such as temperature, pH, nutrients, and salinity helps produce an algal biomass with maximum desired characteristics such as high density or high oil content. Therefore, only a few species that can grow in such selective environments can be grown in open systems.

#### Light

Ample light is required as a primary characteristic of any photosynthetic system. Without light, algae will not be able to convert solar radiation into energy for growth. The surface of an open pond has greater photosynthetic efficiency than photobioreactors because the ponds have more surface area, meaning greater access to sunlight.

Nevertheless, natural illumination presents many challenges to efficient production of algae, such as climatic, seasonal, regional, and diurnal light and temperature variations, as well as the capacity of



**Figure 6:** Light intensity (solid line) and productivity (dotted line) in an open pond at high light intensities. Productivity is highest at the top or surface, where light is most intense. However, if the light at the surface is too intense for extended periods of time, the algae can die. This holds true for open ponds and outdoor closed photobioreactors.

Source: Wageningen University, [http://www.algae.wur.nl/UK/technologies/production/open\\_systems/](http://www.algae.wur.nl/UK/technologies/production/open_systems/)

algal cells to utilize available natural light. Cloud coverage also redirects irradiation, with an adverse effect on biomass productivity.<sup>42</sup>

Mutual shading will influence algal growth rates as well. Paddle wheels help mix algae, balancing their exposure to light. When no mixing occurs, the algae on the water's surface get too much light, causing photoinhibition (limiting growth), and the algae underneath the surface do not get any light, also preventing growth (Figure 6).

In addition, cell division (growth) and lipid accumulation (energy content) in algae are understood to be mutually exclusive.<sup>43</sup> Manipulation of certain biological and/or environmental parameters can help determine which of these characteristics is promoted foremost.

## Water

Open systems can utilize many different types of water, including fresh, brackish, alkaline, marine, eutrophic, or mixed waters. The type of water available may dictate the type of cultivation system implemented, the species of algae cultivated, and the nutrients needed. Using low-quality water for algae cultivation has a few significant benefits, which will be touched upon throughout this report.

Brackish waters or effluent streams utilized for cultivation may vary in quality based on seasonal environmental conditions and the presence of fertilizers, pesticides, metals, and other waste. Variation in water quality may in turn be detrimental to the quality of biomass produced. For instance, algal biomass cultivated from effluents high in heavy metals (where algae uptake these metals) may not be suitable for converting into animal feed. Management (recovery and disposal) of the metal and chemical byproducts will be important.

## Nutrients and CO<sub>2</sub>

Algae rely on several nutrients to prosper, such as nitrogen, phosphorous, and carbon dioxide. Each nutrient is an important component of the algal growth cycle. Eutrophic or mixed waters (such as animal litter, tertiary wastewater, and agricultural or industrial effluents) are rich in nitrogen, phosphorus, and other nutrients and minerals. The use of nutrient-rich water helps algae grow and decreases the need for endogenous nutrient inputs. Conversely, nutrient-depleted water may be used as a control mechanism to prevent contamination and improve lipid accumulation, while simultaneously decreasing cell division.

Cultivation systems that do not utilize wastewater must add nutrients such as phosphoric acid and urea or ammonium nitrate. HRAP raceway and circular ponds utilize paddle wheels for nutrient and gas mixing, as well as for balanced light distribution.

Atmospheric CO<sub>2</sub> is adequate for algae growth in the wild; however, most commercial systems inject air, pure CO<sub>2</sub>, or liquid CO<sub>2</sub> to boost productivity. Additional warm air or CO<sub>2</sub> inputs in cold climate



conditions may also keep open ponds at tolerable temperatures, ensuring algae survival and even a degree of continued cell growth.<sup>44</sup>

### *Environmental Impacts of Open Systems*

Core environmental issues—water, land and soil, biodiversity, air, and energy—are identified here and discussed in an effort to evaluate the scalability of open systems. As these systems are currently the most economical and technologically basic to implement and operate, refinement should capitalize on the system’s environmental benefits while focusing development on mitigating environmental concerns and unknowns.

#### **Water**

Today’s engineered, open cultivation systems require large quantities of water. The water demand for vast ponds creates concern, most particularly where either water reclamation or wastewater treatment is not an integral component

of the cultivation process. How water demand for commercial algae cultivation compares to oil seed crops is unclear; nevertheless, such demands on water could present immense challenges for algae biofuels development, particularly in that the majority of these open systems could be located in water-constrained regions (e.g., the American Southwest). The environmental benefits and concerns related to the use of wastewater for algae cultivation are more clearly outlined in the section on integrated systems.

Another consideration is how great an impact millions of acres of ponds (possibly with ground liners) will have on the water table, groundwater salination, nutrient regulation, and natural runoff to rivers and reservoirs (e.g., proximity to the Colorado River or Lake Mead Reservoir; Figure 7). Similar to the development of urbanized environments, expansive algae cultivation facilities



PHOTO CREDIT: MIKE KRAWCZYNSKI

**Figure 7:** The water table at Lake Mead Reservoir, Hoover Dam, Nevada has declined significantly in recent years.



will create large areas of impervious surface and will most likely capture any stormwater for facility use, effectively decreasing rainfall percolation into the soil, aquifers, and wells. This reduction in recharge can vary greatly in effect and is dependent on the percentage of facility coverage in a recharge area.

Decreasing runoff inputs to regional rivers could lead to severe environmental deterioration as well as increased stress on river- and reservoir-dependent residential communities, many of which have already been facing a decades-long water shortage. Even with the recycling of process wastewater, evaporation will require new inputs of water on a regular basis, especially in arid and semiarid climates.

Conversely, coastal ponds may utilize seawater, which would limit impact on freshwater supplies. Nevertheless, the introduction and continuous cycling of saline water through a naturally freshwater ecosystem will likely have some effect on the immediate environment, certainly in terms of an increased chance for chemical contamination and groundwater salination.

High evaporation rates could influence salinity or nutrient concentration. To prevent salt accumulation, some water needs to be discharged continuously from the ponds—the faster the evaporation, the larger the discharge.<sup>45</sup> The impact of continuous discharging will vary depending on the quality and volume of the water and the manner in which it is discharged, and whether it is reused or released into the local environment.

Maintaining sterility at commercial-scale open pond facilities is a challenge and could have environmental implications where the use of pesticides, chemical sterilization, or extreme culture conditions (such as high pH or high salinity) are adopted.<sup>46</sup>

Because of the lack of available data and the limited scale of development, the long-term impact of commercial-scale open pond cultivation on water resources and ecosystem health remains unclear, but greater exploration of related water concerns is strongly recommended.<sup>47</sup>

### Land and Soil

A major advantage of cultivating algae for fuel feedstock is the high yield per unit of land compared with traditional agricultural crops or grasses yielding such biofuels as ethanol or methanol. However, open pond systems are the least land efficient among existing or concept algae cultivation systems. In addition, open systems with integrated wastewater treatment sometimes require additional ponds for algae maturation or sediment settling (i.e., facultative, stabilization, and settling/draining ponds). In such cases, land use and soil impact may need to be weighed against



**Figure 8:** Open ponds often use pond liners to limit water discharge and prevent contamination of culture medium.  
Source: AgriLife Research Mariculture Laboratory, Flour Bluff, Texas.

the combined benefits of biomass production and wastewater treatment.

Open systems have received much support for their technical simplicity, but cultivating a competitive feedstock in a scalable open pond system could transform perhaps tens of millions of acres of land. The environmental implications of such a transformation of the landscape into million-acre ponds have not been given much needed attention.

For instance, new infrastructure for water lifting to deliver water to the cultivation facility (in cases where the water source is not adjacent to the facility), or in recycling or disposing of used water, could also have environmental impacts similar to those caused by new road construction, such as

increased soil imperviousness, increased runoff, and soil erosion.<sup>48</sup>

Secondly, man-made ponds sometimes feature ground liners (synthetic or natural) to protect both the algae culture and underlying soil from contamination and to prevent process wastewater from infiltrating groundwater (Figure 8). The use of pond liners increases environmental impacts, which will vary depending on the type of liner used and the quantity of ponds in the system. Soil enhanced with bentonite clay is the most affordable solution but requires professional installation. This soil may also absorb compounds from the water and thus may not be suitable for certain environments.

Synthetic membranes are the most effective, but they bring forth other environmental concerns such as the toxicity of the membrane material and recyclability or disposal after its use. Life cycle analyses may be important for identifying which materials will have the least impact under given conditions.<sup>49</sup>

### Biodiversity

Maintaining algae culture purity is very difficult and it also means limiting or excluding natural and native aquatic life. Whereas algal bloom problems in the natural environment are typically due to nutrient imbalance of the water and not necessarily the “escape” of algae, exotic and potentially invasive algal species from engineered cultivation systems may threaten the integrity of local and regional ecosystems, including those downstream from cultivation pond runoff (and harvesting discharge).

System proximity to protected ecosystems (parks, natural reserves, Native American reservations, etc.) may meet resistance from local government or residents.

A consensus has not been reached as to whether the cultivation of genetically modified algae could also risk contamination of native ecosystems (see sidebar on transgenic and genetically modified algae and Appendix B for more discussion on genetically modified algae).

The semiarid, barren expanses in the American Southwest have been suggested as ideal places for

### Transgenic and Genetically Modified Algae

Transgenic algae possess a gene or genes that have been transferred from a different algal species or other organism. Although DNA of another species can be integrated into an algal genome by natural processes, the term “transgenic algae” refers to algae created in a laboratory using recombinant DNA (from a genetically modified organism, GMO) technology for the purpose of designing algae with specific characteristics. In the early 1990s, genetic engineering (or mutagenesis) was determined to be the more promising way “to produce algal strains with constitutively high lipid levels.”\*

There are many factors that, separately or in combination, reduce the overall productivity of an algae cultivation system. The use of transgenic or genetically modified algae in closed (i.e., more controlled) cultivation systems may be able to address a number of biological barriers to high-yield cultivation, including organism survival, growth rate and lipid content, CO<sub>2</sub> absorption rates, light penetration, and temperature, as well as tolerance of high-stress harvesting methods.

In general there are two main concerns with GMO technology: the escape of GMO organisms into the natural environment and the loss of biodiversity brought about by the displacement of naturally occurring organisms. While the cultivation of genetically modified algae for nutraceuticals has been practiced for decades with little or no reported environmental impact, the difference in scale between algae production for nutraceuticals (small scale) and for bioenergy (large scale) is too great to assume an understanding of potential environmental impacts. (See Appendix B for further discussion on the impacts, regulations, and policies associated with genetically modified algae.)

\* Sheehan J., Dunahay T., Benemann J.R. and Roessler P., 1998. A look back at the U.S. Department of Energy’s aquatic species program: biodiesel from algae, Golden, CO, National Renewable Energy Institute, NREL/TP-580-24190:113.

extensive open pond systems, as they would not displace livestock or food agriculture. However, the introduction to this region of thousands of square miles of surface water (akin to the surface area of some of the smaller Great Lakes) could have immense impacts on the desert ecosystem or any

region where such a vast land transformation is implemented.

Negative effects on biodiversity will likely occur, to varying degrees, depending on the location and scale of a cultivation facility. For instance, the migration of birds and terrestrial wildlife may be of very relevant concern. An increase in humidity and overabundance of surface water, or perhaps lack of groundwater, may contribute to loss of habitat and affect bird migration patterns because the natural migratory path will have been altered.

### **Air**

Although high concentrations of algae cells will likely decrease pond evaporation rates, vast commercial-scale open pond cultivation could potentially alter precipitation patterns and native habitats, which could dramatically affect local and regional ecosystems. Although the anticipated increase in regional humidity is at present only theoretical, the implications could be great enough to warrant close consideration in open system planning, design, and localization. Open pond cultivation facilities should consider region-specific pan-evaporation rates.<sup>50</sup>

### **Energy**

In determining the energy balance of open systems, a number of energy inputs must be considered, including culture inoculation, CO<sub>2</sub> injections, nutrient balancing, paddle wheel operation for culture mixing, machinery maintenance and upgrades, and whole-system cleaning. In general, open systems require less maintenance and tend to utilize less energy than most other cultivation systems.

Although CO<sub>2</sub> inputs are not necessary for open cultivation, those systems that incorporate CO<sub>2</sub> inputs do so as a continuous and perhaps energy-intensive process. The use of liquid CO<sub>2</sub> is an additional energy burden as it is delivered to the site after undergoing an energy-intensive pressurized liquefying process.

Because open systems require large expanses of relatively flat land, the integration of artificial CO<sub>2</sub> inputs may only be practical where power plant flue gas is easily accessible and there is a CO<sub>2</sub>

scrubbing option of value, as it would be necessary to rid the flue gases of any heavy metals.<sup>51,52</sup>

Finally, open ponds operate with a variety of hydraulic retention times (HRT) before harvesting. Longer HRTs could mean increased vulnerability to contamination of both the growth culture and the natural environment. Energy inputs may also be greater in colder climates, where warm air is sometimes pumped into ponds to keep the culture at a desired temperature. Improved cultivation conditions would likely improve algae productivity and reduce the HRT, subsequently decreasing energy inputs and risk of contamination.

### ***Open Systems Summary***

There are foreseeable challenges to open pond sustainability, especially where water is scarce and ecosystems are at risk, and where there is no associated (primary or secondary) environmental benefit such as wastewater treatment. Environmental barriers to scalability include the availability of water, water use impact on the greater watershed, transformation of land and soil characteristics, alteration of regional climate (due to heightened evaporation rates), and a decrease in the populations of native flora and fauna.

In addition, million-acre cultivation facilities could make sustaining the water table at a healthy level one of the greatest challenges of commercial-scale algae-to-biofuel production. Therefore, the locating of algae cultivation facilities should take into account the relationship of the facility to the functioning of the greater watershed, including dependent ecosystems (both terrestrial and aquatic) and residential communities, before any commitment is made to commercial-scale open pond facilities.

Maximum culture densities and proportion of oil in the final biomass are essential to measuring scalability of any algae cultivation system. Proximity to harvesting facilities will also be a consideration for determining sustainability because of the environmental implications of biomass storage and transport (many of which are not addressed in this report). Locating cultivation systems near downstream processing facilities would minimize the need for such storage and transport.



### CLOSED PHOTOBIOREACTORS

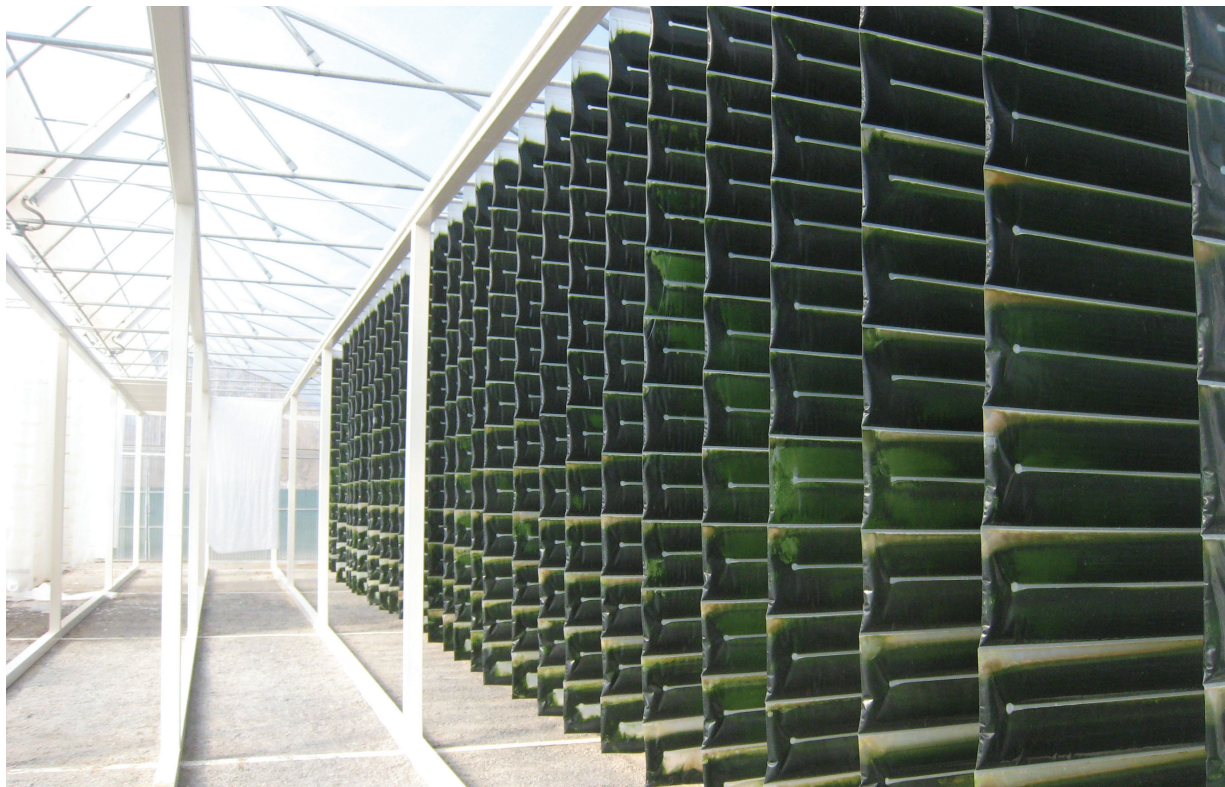
Closed photobioreactors (PBR) are an approach to algae cultivation that aims to overcome many of the biological limitations and environmental barriers faced by open pond systems. PBR systems are more technologically complex compared to open systems. There is some expectation that PBR cultivation could improve efficiency in attaining greater biomass density and provide potential environmental benefits, such as decreased inputs of certain natural resources.

#### *System characteristics*

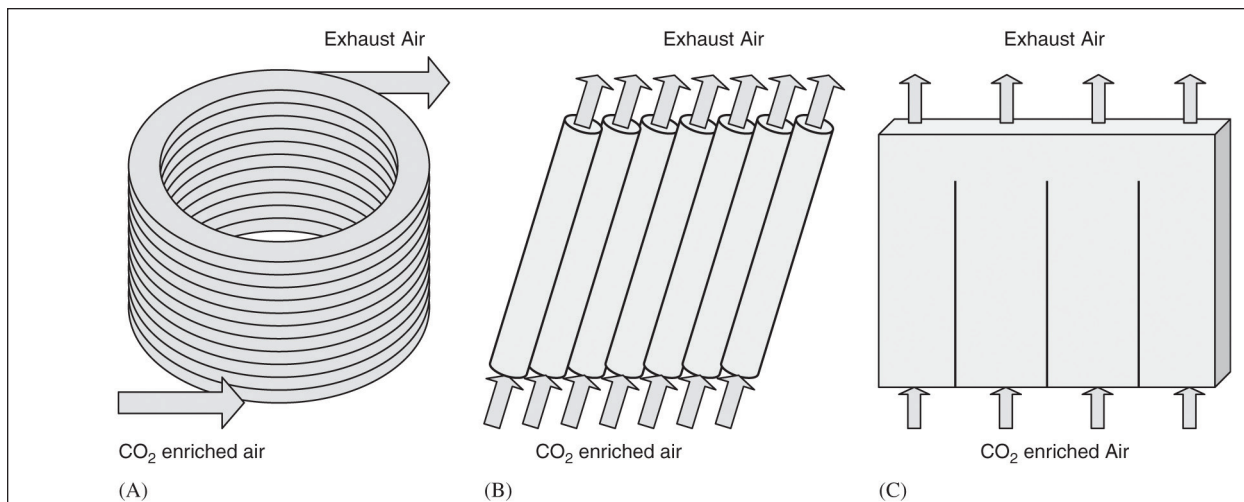
A PBR can be described as an enclosed culture vessel that is designed to utilize light to support photosynthesis for controlled biomass production. Because of the variety of approaches taken to balance light distribution with maximizing culture density and total oil content, countless PBR designs have emerged that can be categorized generally into either indoor or outdoor closed PBRs.<sup>53</sup> Indoor closed PBRs usually require artificial illumination. Outdoor closed PBRs utilize natural daylight and in some cases may also incorporate artificial illumination. PBRs tend to

have higher volumetric productivity than open ponds. The most efficient large-scale PBRs should in theory accommodate large volume, occupy less space, have high biomass yields, and, for outdoor PBRs, should also have transparent and high illumination surfaces.<sup>54</sup>

General categories of PBRs include indoor/outdoor polyethylene sleeves or bags that either hang over land (Figure 8) or float in water; outdoor tubular and flat plate systems (Figure 10) that come in several variations; and indoor columns or modular tank systems (Figure 10; see Pathway Map C-I).<sup>55</sup> Continuous and hybrid PBR systems, which are variations of the linear, single-step cultivation practices discussed thus far, address more specific biological limitations and economic barriers to large-scale algae cultivation (see sidebar on Continuous Production and Hybrid Systems for further discussion). Each of these various PBR systems has its advantages and disadvantages. Compared with open ponds, however, PBRs exhibit better control of temperature, pH, and light intensity, with higher biomass densities in lower quantities of water and on less land.

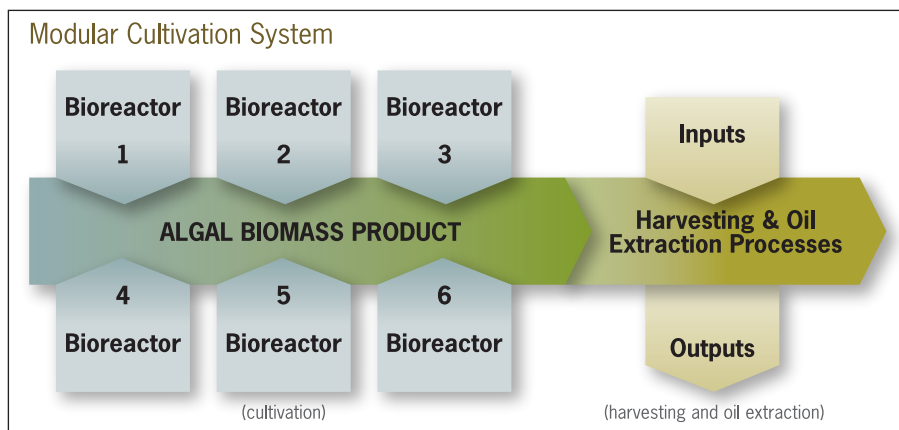


**Figure 9:** Valcent’s High Density Vertical Growth (HDVG) systems grow algae with only light, water, and air in a closed loop, vertical system of polyethylene sleeves in greenhouses.



**Figure 10:** Some examples of translucent photobioreactors fed with air enriched with CO<sub>2</sub> for mass cultivation of algae.

Source: Muñoz and Guieysse, 2006.



**Figure 11:** Modular tank closed photobioreactors combine the mature culture medium before downstream processing. This approach makes modular cultivation systems relatively easy to scale up over time, from one bioreactor to many, tapping into an established infrastructure and requiring only relative scale-up of harvesting and oil extraction machinery and capacity.

In theory, PBR design can be manipulated and reengineered to account for different regional environmental parameters in which the system is developed, including land and climate conditions, structural characteristics, preferred algal strain, intended productivity, energy flows, and associated products. Because of this wide variety in PBR designs, the algal species and necessary light, water, and nutrient inputs tend to vary from system to system. Discussed here are the general characteristics of PBR inputs.

### Algae

Closed PBRs tend to be designed to support algae monocultures; there is a range of species, both natural and transgenic, currently being used.<sup>56</sup> One of the primary operational disadvantages of closed PBRs is the propensity for algal film buildup.

Some species of algae will grow on the inside surface of a PBR, prohibiting light from penetrating further into the PBR. Unless the species selected is known to not grow on PBR surfaces, either routine system cleaning or the application of a special coating to the inside of the PBR will be required.<sup>57</sup>

### Light

Efficient light delivery and distribution are principal obstacles to using commercial-scale PBRs for algae cultivation and are thus stressed as primary design characteristics in PBR innovation. Tubular PBRs, for instance, tend to have higher light utilization efficiency than flat-plate systems because of larger reactor surface area per unit of land.<sup>58</sup>



## Production Modes

### Continuous Production

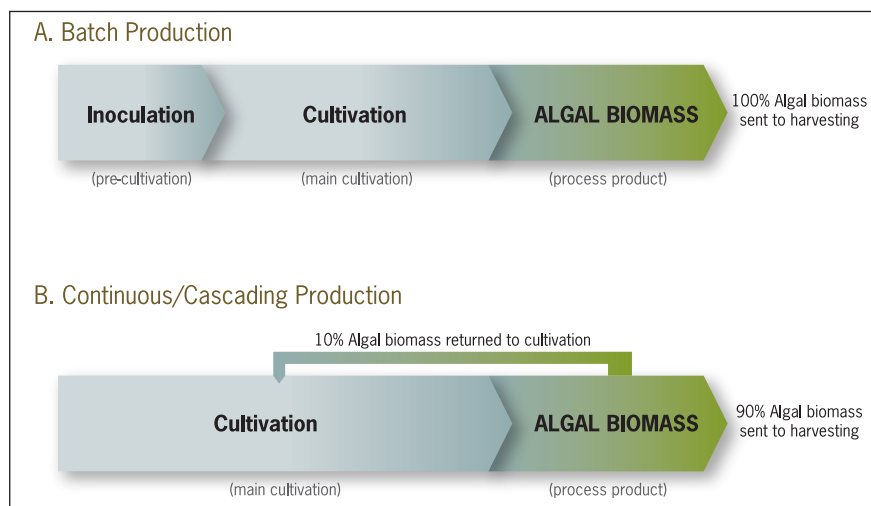
Continuous production (also known as cascading production) aims to improve process efficiency, whereby a percentage (~10 percent) of the harvestable biomass is returned to the cultivation platform (i.e., PBR) to inoculate the culture medium for cultivation of the next “batch” of biomass, while the remaining (90 percent) biomass is harvested (Figure 12). This allows the cultivation process to continue with less frequent system shutdowns for cleaning and inoculation compared with a PBR that does not incorporate continuous production.

### Hybrid Systems

Since most algae do not grow simultaneously by cell division and lipid accumulation—generally, they are mutually exclusive measurements of productivity—a hybrid cultivation process may be employed to first increase culture concentration, then to increase lipid accumulation. This two-step (closed-to-open)

cultivation process is a hybrid solution addressing the benefits and limitations of both closed PBRs and open systems.

In the first step, large, nutrient-rich inoculum of anoxic algae is produced in a closed PBR, which promotes cell division and minimizes chance of contamination. The second step is typically conducted in raceway HRAPs, in continuous mode with low nitrogen content, to promote the biosynthesis of algal oil.\* The expected result is high biomass yields with high oil content. Ultimately, these yields will determine scalability of hybrid systems if they are able to maximize resources and improve efficiencies while decreasing overall environmental impact. Alternatively, Muñoz and Guieysse (2006) suggest developing new treatment methods such as “membrane photobioreactors” or “combined physical-biological processes” to improve biomass control and protect algae against inhibitory effects.



**Figure 12:** Unlike batch production (A), continuous production (B) allows for near continuous cultivation and reduces system maintenance.

\* Hu Q., Sommerfeld M., Jarvis E. and Ghiarardi M., 2008, “Microalgal triacylglycerols as feedstocks for biofuel production: perspectives and advances,” *The Plant Journal* 54:635; Vasudevan P.T. and Briggs M., 2008. “Biodiesel production—current state of the art and challenges.” *Journal of Industrial Microbiology and Biotechnology* 35:428.

Some PBR systems use a combination of natural and artificial light sources. Artificial lighting (e.g., fluorescents, LEDs) can be provided externally or from within the PBR, such as with light rods inserted into PBR tanks. Natural light can be provided through the application of translucent materials or remote solar lighting systems meant to enhance sunlight utilization.

### Water

As with open ponds, some PBR systems can be designed to accommodate eutrophic or tertiary wastewater. Although contamination of algal cells in closed PBRs is of lesser concern than in open systems, any nonsterilized water source may carry viruses or predatory organisms that could pose a threat to biomass integrity.

Unlike open systems, closed PBR systems can be designed to provide external heat exchange for temperature control and to minimize evaporation. Water temperature is also more easily controlled in indoor PBRs than in outdoor PBRs or open systems located in cold climates, supporting a longer (year-round) cultivation season.

#### **Nutrients and CO<sub>2</sub>**

Nitrogen and phosphorous not provided by the water source must be added to the culture medium, in the form of phosphoric acid and urea or ammonium nitrate, on a regular basis. Since closed PBRs are not designed to utilize atmospheric CO<sub>2</sub>, a continuous artificial supply of soluble inorganic carbon will also be required to support algae growth.<sup>59</sup>

Effective injecting and mixing of the CO<sub>2</sub> and other nutrients in the culture medium is likely to be of critical importance to ensuring culture stability. Gas transfer is an obstacle for closed PBRs. As photosynthesis occurs, oxygen increases; open ponds utilize paddle wheels to support gas transfer, whereas PBRs require the application of a degasification system.<sup>60</sup>

#### ***Environmental Impacts of Closed Systems***

Environmental issues concerning the scalability of closed PBRs are discussed here in terms of water, land, soil and biodiversity, air, and energy, and a sidebar addresses materials toxicity. In general, many of the environmental impacts of closed PBRs are similar to those of open systems. The degree of impact may vary; however, in many cases, data on core issues are too limited to state decisively.

#### **Water**

As with open ponds, the water source may be site specific (i.e., based on water type and availability) and may also depend on restrictions set forth by local or regional water rights and administrative regulations. Likewise, the impact of water use by closed PBR systems will depend on the management of downstream (i.e., postcultivation) discharge of process wastewater. Depending on PBR design (and harvesting techniques employed), process wastewater could be easier to recover, treat (if necessary), and recycle through the cultivation system.

Although PBR systems are not subjected to variables like rainfall and evaporation, the water demand will likely be high and of great importance to the sustainability of the facility and the ecosystem from which the water is withdrawn. Water-related impacts of closed PBR cultivation will be more directly associated with the water source than with the impact of the facility, such as with expansive open ponds. This is especially true where water is continuously extracted from ground sources (aquifers) and released (postharvesting) into surface waters that do not replenish the same aquifer from which the water was originally withdrawn.

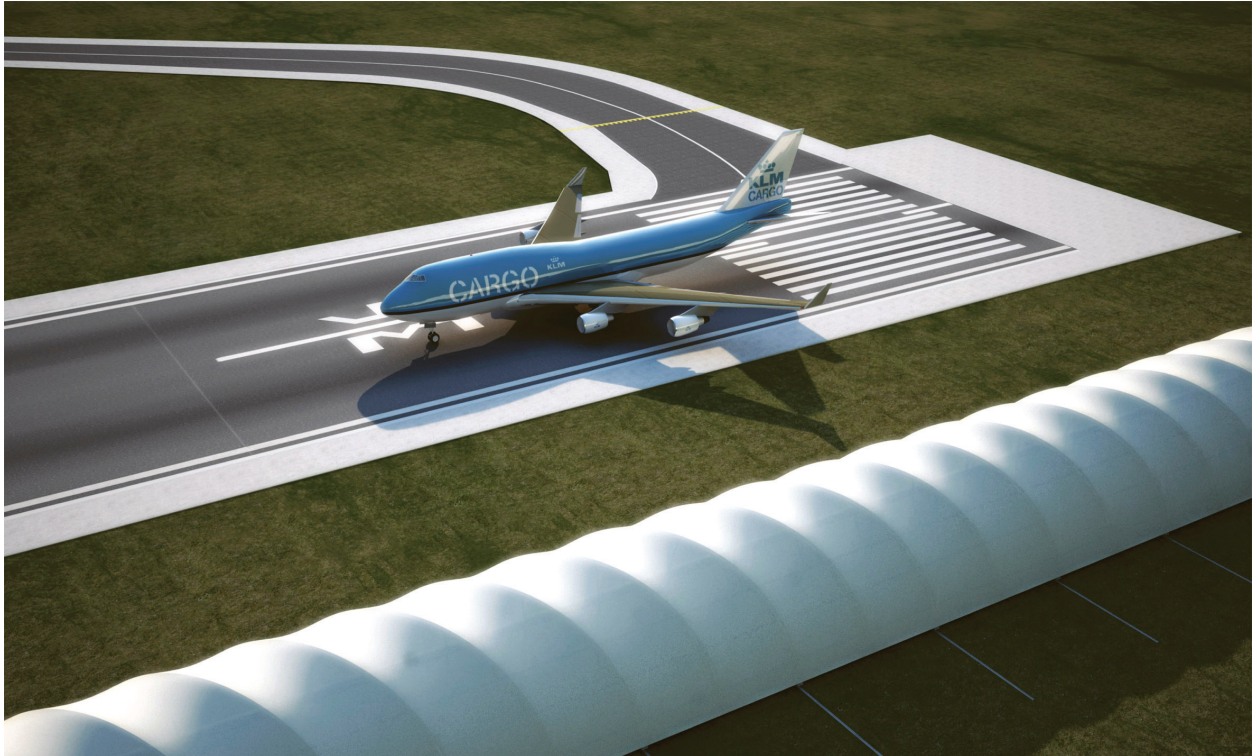
With respect to hybrid systems, water-related concerns will likely mirror those of open systems to some degree, as they are likely to require extensive use of open ponds, lined with impervious ground covers, and accommodating water with highly regulated environmental parameters (e.g., pH, nutrient levels). For both closed PBRs and hybrid systems, sustainable management of water inputs and outputs—source water and process wastewater, as well as stormwater and runoff from production facilities—will be of vital importance.

#### **Land**

Because of the variation in PBR designs, there exists a degree of flexibility in land use. Thus, the quantity and efficiency of land coverage for a closed PBR facility will vary depending on PBR design.

As algae cultivation in closed PBRs (particularly indoor closed PBRs) is relatively independent from regional climate conditions, site location for cultivation facilities is not restricted to particular regions of the country, such as with open pond cultivation. Instead, land use may be defined by proximity to a water source and to downstream processing facilities, which would determine the need for water lifting and eliminate the need for biomass storage and transport, respectively, while reducing overall product production time.

However, outdoor PBR systems could be more land intensive than indoor PBRs because of their need for optimizing access to daylight.



**Figure 13:** Schiphol Airport, Holland. Construction of an “ecobarrier” is meant to protect a neighborhood close to the airport runway from low frequency noise. The barrier is a tent, constructed parallel to the runway, open for noise waves on one side and closed on the other side. The ecobarrier supports algae cultivation and biofermentation technologies, essentially integrating one component of transportation infrastructure into the landscape of another.

Source: Aviationwatch.eu

Some designs (polyethylene bags, in particular) do have the advantage of vertical arrangement and increased area for natural light inputs. More compact and vertical designs would improve the ability of facilities to be located in urban districts.

Other PBRs are designed to be placed on unoccupied rooftops (small scale) or interspersed with other technologies, such as photovoltaic panels, wind turbines, or other infrastructure (Figure 13), allowing maximum utilization of available land. Such integration of technologies may address one of the primary concerns with open systems and has the potential to make a significant contribution to land use efficiency.

Given the cost and availability of land in the United States, increasing algae productivity could effectively decrease acreage of land needed as well as decrease impact on soil quality and regional ecosystems.<sup>61</sup> Modular PBR systems that can be scaled up over time, from one bioreactor to many, could require only relative scale-up of harvesting

and oil extraction machinery and capacity, which could present a more sustainable use of land when developing a commercial-scale cultivation system.

### Soil and Biodiversity

Contamination and soil conditions (including impermeability) by commercial-scale closed PBR facilities could become environmental issues so far as building construction and systems discharge are concerned.

Although the cultivation of algae in closed PBRs may not pose a direct threat to soil quality and biodiversity, the process wastewater released into the natural environment may carry metals, chemicals, nutrients, or nonharvested algal cells. On the chance that the cultivated algae were an exotic, invasive, or modified species, the natural environment could be affected by exposure to algal blooms, ecosystem dominance over native species, or contamination of organic food crops. There also exists the potential for environmental

contamination caused by spillage from alternative cultivation pathways such as hydrocarbon-excreting algae. These “renewable oil spills” could contaminate nearby soil and waterways.

Although these scenarios are not probable, the management of cultivation pathways and process wastewater will determine their likelihood.

#### Air

Closed PBRs can recycle industrial CO<sub>2</sub> emissions by injecting CO<sub>2</sub> into the culture medium. However, industrial emissions will need to be cooled and there may also be hazards associated with tapping industrial plants such as those concerned with concentrating heavy metals. Nevertheless, CO<sub>2</sub> is a required input for closed PBR cultivation and some solution will be needed.

In addition, the increase in energy demands for closed systems could increase the life cycle carbon emissions of this process, depending upon the carbon intensity of the type of energy utilized (e.g., coal/natural gas or renewable electricity).

#### Energy

While PBRs have the added benefit of parameter control mechanisms, automation, and potentially decreased labor and maintenance, they also demand continuous light energy that, from an energy consumption standpoint, could prove impractical for the energy balance of fuel production.

Open ponds and outdoor (translucent) closed PBRs are driven by natural light, the availability of which remains unchanged with scale-up, whereas the scale-up of indoor closed PBRs requires an increase in artificial light, which could be very energy intensive. Continuous delivery of artificial lighting via electricity will have a significant impact on the energy balance, and potentially the life cycle carbon emissions, of commercial-scale production.

Unlike open systems, PBR design is closely linked to energy inputs. Efficient light distribution often demands larger surface areas or improved light dispersal through a denser culture. Greater surface area means more materials (for PBR fabrication) and increased land area. Achieving higher densities

### Materials Toxicity and Pathway Energy Balance

Fluorescent bulbs, LEDs, or other artificial light sources are typically used to provide the irradiance required to sustain photosynthesis of photoautotrophic algae for small-scale, indoor closed PBRs producing high-value products. This scenario may prove to be environmentally costly when scaled to commercial biofuel production.

Where artificial light inputs are particularly high, the environmental impacts of bulb materials will need attention. Fluorescent bulbs are made with some mercury and LEDs have other heavy metal toxins. Although the quantity of mercury may seem dismissible on a system basis, given the quantity of bulbs required to sustain commercial-scale cultivation in closed PBRs, disposing of retired bulbs could become a contaminating process. Bulbs should be designed with minimal heavy metals and fewer Watts per lumen.

PBR systems designed with transparent surfaces typically use materials such as PVC, Plexiglas, or glass. This is important to note when measuring the energy required for mass-production of PBRs. Producing glass is an energy-intensive process; glass PBRs can fracture or break yet can also be recycled. Plexiglass is more durable, but is also a petroleum-based product that is not easily recycled. Polyvinyl chloride, or PVC, is cheap, durable, easily recycled, and thus a widely used thermoplastic polymer; however, the use of PVC in consumer packaging is known to pose threats to human and environmental health and negatively affect the recycling stream.\*

The toxicity of pond liners for open pond and integrated cultivation systems could also pose a threat to biodiversity, soil and water quality, or aquifer recharge. The environmental impacts of materials toxicity and energy inputs for materials fabrication will be important considerations for measuring the sustainability of any biofuel production pathway. Environmentally responsible materials should therefore be explored in more depth and, ideally, designed to exceed the latest regulatory demands placed on industry.

\* The State of California is currently considering a bill that would ban the use of PVC in consumer packaging because of the threats it poses to human and environmental health and its effect on the recycling stream. The bill will prevent human and environmental exposure to toxins, as well as encourage the recycling of consumer packaging, by phasing out the use of toxic, nonrecyclable PVC packaging. According to the bill, PVC packaging is a threat to human health and the environment. PVC packaging is toxic in all stages of its life cycle, including production, which involves large amounts of chlorine gas and vinyl chloride, a dangerous carcinogen. Studies of PVC have linked it with high cancer rates; [www.cawrecycles.org/issues/current\\_legislation/ab2505\\_08](http://www.cawrecycles.org/issues/current_legislation/ab2505_08)



also requires increased HRTs for what may only be small gains in biomass. The denser the growth culture, the more difficult light dispersal becomes. Therefore, water volumes, lighting, HRTs, and materials fabrication need to be weighed against associated energy demands.

**Closed Systems Summary**

Foreseeable environmental barriers to scalability include the availability of water, water use impact on the sustainability of the greater watershed, potentially high electricity inputs, and implications of unsustainable materials use.

Biomass densities achieved are a driving factor in determining system scalability. With this in mind, a closed PBR could prove more efficient than an open pond because it achieves higher densities year-round. However, very few large-scale closed PBR systems have been implemented, so the feasibility of optimal unit sizes and efficient light delivery and distribution is

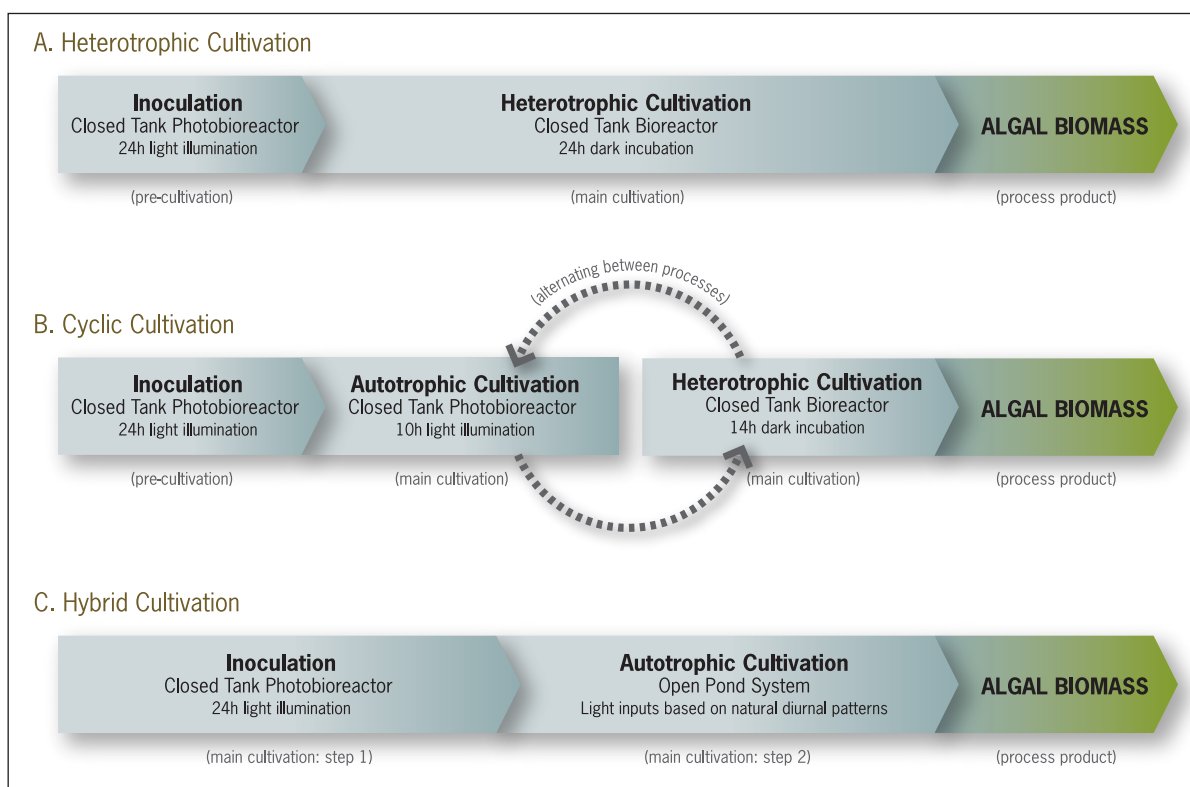
still difficult to ascertain. Although outdoor, vertical PBRs may be a space-efficient alternative to artificially illuminated indoor PBRs, the added expense for transparent materials could make scale-up financially and environmentally burdensome.

**HETEROTROPHIC FERMENTATION**

Heterotrophic fermentation, also known as dark feeding, is an alternative approach to algae cultivation that is fairly well established in some industries. Heterotrophic fermentation is a measurably different approach than open ponds or closed PBRs.

**System Characteristics**

Whereas open ponds and closed PBRs appear more in recent literature, heterotrophic fermentation has already achieved economic efficiency for certain high-value products serving the health and pharmaceutical industries, as well as for some low-value consumer products, such as laundry detergent and carpet fiber.



**Figure 14:** Cyclic cultivation combines technologies of both heterotrophic systems (A) and closed photobioreactors. Cyclic cultivation differs from hybrid cultivation in that its two methods are applied in a continuously alternating loop (B), whereas the two methods in hybrid cultivation are applied contiguously (C), but not in a repeating loop.



Heterotrophic systems are similar to closed PBR cultivation in that both pathways can utilize conventional closed bioreactors, such as stainless steel tanks.<sup>62</sup> The defining characteristics are that high-density heterotrophic cultivation is achieved with inputs of an organic substrate (sugar) feedstock in a zero-light, low-moisture environment (see Pathway Map D-I).

Variations in heterotrophic cultivation are known to incorporate some photosynthetic cultivation practices, but these are not common. Cyclic cultivation is one such example that alternates internal illumination with dark incubation (Figure 14).<sup>63</sup>

Heterotrophic fermentation systems require varying levels of inputs. As discussed here, algal strains, feedstocks, and water inputs tend to be site specific.

### **Algae**

Heterotrophic systems cultivate heterotrophic algae in high concentrations in the absence of light. Heterotrophic algae are photosynthetic organisms with heterotrophic characteristics, meaning that they have the capacity to grow in the absence of light provided that other conditions are met, such as accessibility to organic substrates. The input of organic substrates may increase risk of culture contamination and need for sterilization; however, sterilization of inoculum by steam may eliminate such risk.

Researchers have been able to genetically modify particular algal species to thrive on organic substrates in the absence of light.<sup>64</sup> Because of the variety of naturally occurring feedstocks allowing for flexibility in algal species selection, such species modification may not be a prerequisite for efficient heterotrophic cultivation.

### **Feedstock**

Unlike open pond or closed PBR systems, heterotrophic fermentation requires an organic substrate feedstock (e.g., cellulosic sugars from switchgrass, sawdust, sugarcane, waste glycerol, sugar beet, cellulosic pulp, low-grade molasses) as a carbon source for the algae to grow. Organic carbon can be delivered at regular intervals in the form of glucose, sucrose, acetate, fructose, or ethanol. The utilization rate of feedstocks will vary depending on the algal species and

conditions of the culture medium.<sup>65</sup> Likewise, the algal species selection may be based upon feedstock availability.

### **Water**

Organic substrate inputs tend to have high moisture content. Combined with recycled system water, water inputs for heterotrophic systems could be kept to a minimum. Wastewater streams may also be able to provide some nutrients. The quantity of water required to operate a heterotrophic cultivation system has not been easy to identify.

### ***Environmental Impacts of Heterotrophic Fermentation***

Environmental issues concerning the scalability of heterotrophic cultivation systems are discussed here in terms of water, land, and energy. Although these issues are essentially the same as those for other cultivation pathways, the potential scale of impact is more difficult to ascertain with heterotrophic fermentation because of the origin and diversity of indirect water, land, and energy inputs (i.e., for growing organic substrates).

### **Water**

Heterotrophic fermentation has a low water demand relative to other cultivation systems. This low demand decreases the need for water management, including delivery, sterilization, recovery, treatment, and release. However, much of the water burden is likely to be shifted from biomass production to the cultivation of organic substrates (in the form of corn, sugar beet, sugarcane, etc.). The use of organic substrates originating from irrigated, nonwaste feedstock suggests indirect water use by heterotrophic systems could be significant at commercial scale.

### **Land**

Land use issues for heterotrophic cultivation are similar to those for indoor closed PBRs. Since heterotrophic systems are typically comprised of steel tanks, climate is of lesser relevance and land use will be minimal compared to open systems; however, the space needed for artificial illumination has been eliminated.

System dependence on organic substrates also has potential environmental implications depending on the source and quantity needed. There is some flexibility in sugar sources (both five- and six-carbon sugars). Some sources must employ arable cropland, while others could be derived from municipal, agricultural, or industrial waste, such as waste glycerol from biodiesel production facilities.<sup>66</sup> This could come at the cost of feedstock storage and transport with minimal impact to land, soil, and water, while decreasing the need for municipal waste disposal. Locating fermentation systems near available feedstock would minimize environmental damage associated with feedstock transport and storage.

### **Energy**

Direct energy inputs for heterotrophic systems are likely to differ dramatically from other cultivation systems. As with open systems and closed PBRs, aeration and mixing in heterotrophic systems require energy. However, in addition to having high culture densities, low water demand, and no light inputs, heterotrophic systems consequently reduce energy requirements for separating cells from water at later stages in the production process.<sup>67</sup> Algae generate their own heat when they are metabolizing, bringing about an incremental decrease in energy use and reducing demand for heating the fermentation tanks in cold climates.<sup>68</sup>

By contrast, the input of organic feedstocks assumes additional processes for cellulose breakdown. Before administering organic substrates to the culture medium, cellulosic feedstocks must first be processed through hydrolysis (such as with first-generation biofuels).<sup>69</sup> However indirect, this energy-intensive process could affect the energy balance of heterotrophic cultivation.

### ***Heterotrophic Systems Summary***

Heterotrophic systems are theoretically easier to scale up than other cultivation systems because water and light inputs do not impose the system design challenges (and thus financial challenges) they do with closed PBRs. The primary challenge to scalability for heterotrophic systems is the potential environmental cost and seasonality of organic substrate

feedstocks. Readily available, year-round feedstocks would be preferred to imported feedstocks; however, heterotrophic systems can be scaled based on a particular feedstock and its availability. Where glycerol is used as the organic substrate input, cultivation can be scaled to whichever strain of glycerol is available (e.g., from a nearby biodiesel plant). At a minimum, the sustainability of a particular feedstock will depend on water and land use (including for storage), processing inputs, and seasonal availability.

If environmental concerns can be addressed and energy balance can be achieved, heterotrophic fermentation may become a particularly attractive pathway in situations with no access to light inputs; in more urban, cold climate environments with land limitations; or where adequate sources of inexpensive organic substrates are readily accessible year-round.

### **INTEGRATED CULTIVATION SYSTEMS**

Integrated cultivation systems combine biological wastewater treatment with algal biomass production. This approach differs from other cultivation pathways in its ability to function as a multipurpose system for such processes as bioremediation, biological wastewater treatment, methane and biofertilizer production, nutrient recycling, sustainable energy, and carbon management. The capacity of certain algal species to absorb or “fix” nutrients, carbon dioxide, metals, and other contaminants have made algae the focus of much discussion about targeting efforts to reduce atmospheric CO<sub>2</sub> concentrations from fossil fuel use.

Although not currently employed for their capacity to accumulate high quantities of algal biomass—where the biomass is harvested and is not beneficially used—such systems could theoretically serve as a feedstock source for biofuels production.<sup>70</sup>

### ***System Characteristics***

There is potential for great diversity among integrated systems. Discussed below are biofilm processing, periphyton filtration, aquaculture, algal turf scrubbers, wastewater stabilization ponds, and integrated wastewater treatment. These systems typically engage heterogeneous algae to naturally purify effluent streams.

### Biofilm Processing

Biofilms can be grown in ponds or photobioreactors and essentially function as fermentation converters yielding methane biogas.

### Aquaculture

Algae-based aquaculture systems, developed to provide nutrients for commercial fish hatcheries, have the added benefit of purification of effluent water from intensive fish production.<sup>71</sup>

### Algal Turf Scrubbers

The algal turf scrubber (ATS®) is an outdoor technology that uses algae to treat polluted waters in either eutrophic waterbodies, shallow (a few centimeters deep), or sloped (raceway) ponds (Figure 16; see Pathway Map E-I).<sup>72</sup> ATS systems rarely use mechanical actions to create the wave or surge needed to mix the culture. The surge is instead created by a pulsing release of flow, which does require a small amount of energy, but is essentially part of the pumping process. Whereas algae cultivation is enhanced by the flow and surge produced by this pumping, in some river systems such as with the ATS system in the Susquehanna River, no pumping is needed.<sup>73</sup> The ATS system can yield a high-quality biomass.<sup>74</sup>

### Periphyton Filtration

Periphyton filtration is a technology similar to the ATS system for performing bioremediation of polluted water. The filters on which the algae grow have a short HRT and the systems can be very large in scale and constructed from harvestable modules.

### Waste Stabilization Ponds

Waste stabilization pond (WSP) systems are much like open systems, but with the primary purpose of treating waste. Much like other integrated



**Figure 15:** A 2.5-acre algal turf scrubber can provide nutrient control for over 10 million gallons per day.

Source: HydroMentia (2008)

systems with the potential to generate bioenergy, WSPs have the advantage of operating with low (or zero) energy requirements. Conventional WSP technology utilizes sunlight to disinfect wastewater without the need for chemicals or electricity consumption.<sup>75</sup>

### Advanced Integrated Wastewater Pond System

The advanced integrated wastewater pond system (AIWPS®) has been used to describe integrated wastewater treatment processes that utilize fermentation and photosynthetic oxygenation to treat sewage and organic industrial wastes.<sup>76</sup> The application of an AIWPS can vary, including aquaculture, municipal wastewater treatment, and de-eutrophication of waterbodies.

An AIWPS typically utilizes raceway configured HRAPs or extensive, open multipond layouts similar to open systems, with paddle wheels for mixing to support low-maintenance wastewater treatment.

### System Parameters

The growth culture of these integrated platforms shares similar inputs with open pond systems, but at a level more tolerant of natural variables such as algal species, light, nutrients, and other parameters. Accordingly,

they can operate in deeper symbiosis with their natural environment compared to other cultivation systems.

**Algae**

Integrated systems typically support heterogeneous algal-bacterial consortia. ATS and periphyton filter systems are usually colonized by wild, filamentous algal species native to the immediate area or region and that preexist in the targeted water system.<sup>77</sup> In this environment there is low risk of unnatural contamination, although a pesticide may be needed to control insect populations in some instances.

The assemblage of freshwater bacteria, fungi, and periphyton algae (also known as benthic algae) is determined by a number of environmental parameters of which irradiance, temperature, current, nutrient levels, and the degree and frequency of disturbance (i.e., tides, floods) are key.<sup>78,79</sup>

Depending on the purpose of the system, there could be reason to use algal species with particular nutritional profiles.<sup>80</sup> As integrated systems are designed to harness the benefits of native species, GMOs have not traditionally been used in these systems.<sup>81</sup>

Instances where suitable (native) species could be limited and the oil content of those particular species happens to be low, the biological service provided by the algae (e.g., wastewater treatment) is likely to be the primary benefit of the system, while the biomass (residue) is the primary bioproduct and the oil is a value-added benefit.

**Light**

Integrated systems are amenable to seasonal and diurnal light patterns. No inputs of artificial light are necessary and, in many cases, artificial

light may not even be feasible. In colder climates, however, outdoor systems (as most are) may have longer processing times because of limited daylight and low temperatures.

**Water**

Water sources for these systems are existing polluted surface water (e.g., lakes, rivers); or waste streams such as from agriculture, livestock and municipal wastewater; or coal, pharmaceutical, and aluminum plating plant effluents. Although ATS and periphyton filters typically treat eutrophied or polluted freshwater or tertiary wastewater, some AIWPS designs can also accommodate sewage and organic industrial wastes.<sup>82</sup>

**Nutrients**

Wastewater rich in nitrogen, phosphorus, and other nutrients is often well suited for mass-cultivation of algae;<sup>83</sup> this simplifies the function of integrated systems where exogenous nutrient inputs are unnecessary.

In wastewater-fertilized systems, the role of algae is primarily to produce oxygen (O<sub>2</sub>). Mixing of water, nutrients, and gases can be provided by wave simulation, flow release, or pumping. Nutrient uptake, heavy metal absorption, and disinfection are added benefits.<sup>84</sup>

**Environmental Impacts of Integrated Cultivation Systems**

Microalgae have received increased attention recently for their capacity to use resources not suitable for agriculture, including wastewater, marginal or underutilized land, nutrients, and carbon dioxide. Integrated systems capitalize on these characteristics and, as a result, generally require fewer inputs (e.g., nutrients, chemicals); need less energy, operation and maintenance than mechanical wastewater treatment

Table 2. Sample of Algal Species with pollutant fixation capabilities	
Algal Species	Toxin Fixed / Pathogen Inactivation
<i>Chlorella</i> spp.	copper (varies with pH level)
<i>Dunaliella tertiolecta</i>	aluminum plating and pharmaceutical plant effluents
<i>Scenedesmus abundans</i>	cadmium, copper
Algal-bacterial (heterogeneous) consortiums	<i>E. coli</i> and other pathogens



systems; require less land and produce fewer odors; and have a longer life cycle than ordinary algae-based wastewater treatment ponds.<sup>85</sup>

Environmental issues concerning the scalability of integrated platforms are discussed in terms of water, land and soil, biodiversity, and energy.

### **Water**

A prime advantage of integrated systems is their ability to treat polluted water without diminishing groundwater resources. The advantages of wastewater treatment by algae are characteristic of any cultivation system that utilizes polluted waters. Unlike other systems discussed in this report, however, wastewater treatment is the primary function of integrated systems. Integrated systems have higher rates of nutrient recovery compared with other systems and may even improve environmental conditions where the quality of surface water is at risk of causing soil contamination or eutrophication.

Algae effectively remove nitrogen, phosphorus, trace heavy metals, and other contaminants from eutrophied waters or wastewater streams without the use of chemicals (Table 2), essentially leading to a reduction in both pollution and water treatment costs.<sup>86</sup> According to a report prepared for the International Network on Biofixation of CO<sub>2</sub> and GHG Abatement with Microalgae, waste streams from about 30,000 people, or about 5,000 pigs, or 1,200 dairy cattle could be treated economically by approximately 25 acres of algal ponds.<sup>87</sup> Processed water from ATS and periphyton systems is of high quality and can usually be employed for irrigation or released downstream.

The presence of heavy metals in the water source may also dictate the biofuel conversion processes used as well as the end product application. Algal biomass cultivated in the presence of metal toxins is unlikely to be converted into animal feed.

When integrated systems are implemented in place of conventional wastewater treatment plants, the environmental value of nutrient removal and oxygen injection could reduce sewage sludge disposal and increase the quality of water outputs.<sup>88</sup> The use of the biomass in the

production of biofuel (or other bioproduct) would be a value-added benefit.<sup>89</sup>

Additionally, ATS production of “one unit dry weight of algae is known to be accompanied by the release of one and a half times as much dissolved molecular oxygen.”<sup>90</sup> In other words, algae have the capacity to restore water to healthy O<sub>2</sub> levels. Injections of O<sub>2</sub> into a waterbody make the algae-based approach to wastewater treatment particularly attractive for its potential to maintain aquatic health or even recuperate “dead zones.”<sup>91</sup> Additionally, ATS systems temporarily redirect and treat surface water rather than deplete groundwater sources. These characteristics may also be possessed by other AIWPS platforms or any cultivation system that cycles reclaimed water or wastewater rather than extracting water from fresh groundwater sources.

With respect to hybrid systems, water-related concerns will mirror those of open systems to some degree, as they are likely to require extensive use of open ponds that are lined with impervious ground covers and accommodate water with highly regulated culture growth parameters (e.g., pH, salinity, nutrient levels).

### **Land and Soil**

Land-based systems are designed as shallow raceway ponds only a few centimeters in depth. Like open system HRAPs, some land-based ATS systems use pond liners (epoxy-coated plywood and fiberglass for smaller systems and soil bed liners [HDPE] for larger systems) that may affect the immediate soil conditions because of increased imperviousness.<sup>92</sup> Water-based systems that use screens or filters in preexisting waterbodies are less likely to have such impacts on land and soil conditions. Nevertheless, the effect on water surface area of integrated systems should be explored for potential commercial-scale environmental impacts.

### **Biodiversity**

Since the algae employed in these systems are typically of wild, preexisting heterogeneous consortia that are native to the local ecosystem, there is little risk of contamination or negative influence on the natural environment. In fact,



## Summary of Environmental Issues Related to Algae Cultivation

There are many environmental issues with which sustainable algae cultivation will likely be challenged and impacts could vary drastically from system to system. At a minimum, the criteria for sustainable cultivation should consider the effect of water and land usage and potential GMO impacts on biodiversity and ecosystem health, as well as the environmental impacts of infrastructure fabrication, installation, materials toxicity, electricity demands, and waste treatment. Summarized below are the main environmental benefits, concerns, and unknowns.

### ENVIRONMENTAL BENEFITS

- ▶ Minimal competition with food crops for arable land
- ▶ Potentially uses less water than land-based fuel feedstocks
- ▶ Biofixation/bioremediation: economic wastewater treatment and improved water quality
- ▶ Potential prevention and mitigation of eutrophication/dead zones
- ▶ Biomass productivity per unit of land
- ▶ Use of industrial CO<sub>2</sub> emissions

### ENVIRONMENTAL CONCERNS

- ▶ Maintaining sustainable water levels (surface water as well as shallow and deep aquifers)
- ▶ Water quality affects biomass quality; low-quality biomass may require more processing for upgrade, leading to increased energy inputs and undesirable byproducts
- ▶ Competition with food crops for arable land could arise with certain systems

- ▶ Extensive land transformation by certain cultivation systems could alter native habitats and migratory patterns
- ▶ Exotic, modified, or invasive algal species may threaten the integrity of local and regional ecosystems and organic agriculture
- ▶ Electricity inputs could severely affect energy balance
- ▶ System designs may include materials (translucent materials, lightbulbs, pond liners) with high potential for environmental contamination (or other life cycle impacts)
- ▶ Identified environmental impacts may be accepted as trade-offs for more efficient cultivation methods

### ENVIRONMENTAL UNKNOWNNS

- ▶ Water demand (direct and indirect)
- ▶ Effect of water demand on land, ecosystems, and the greater watershed
- ▶ Impact of released/escaped GMO or exotic algal species on the natural environment
- ▶ Energy demand
- ▶ Potential for CO<sub>2</sub> abatement
- ▶ Potential for environmental contamination because of spillage from alternative cultivation pathways, such as hydrocarbon-excreting algae, creating “renewable oil spills”

unlike most other approaches to cultivation, aquatic life is an essential component of integrated systems; therefore, symbiotic organisms are welcomed and natural predators are not perceived as a threat to the system or product quality.

Nevertheless, some aquatic life could be affected either by the insertion of screens and filters into existing waterbodies, or by the diversion of water from the natural flow of a river. The degree of impact, if any, will likely depend on seasonal

fluctuations in water levels and how well the water is managed, such as the rate at which large quantities of water are diverted (upstream) and then reintroduced (downstream).

De-eutrophication, caused by oxygen injections and uptake of nitrogen by the algae during cultivation, will improve aquatic habitat conditions and reduce the likelihood of a waterbody becoming a “dead zone.”

**Energy**

The primary energy benefits of many integrated systems are low maintenance requirements, minimal energy inputs and wastewater treatment, and zero waste byproducts. System operation takes place at ambient temperature, so there is no need to heat or cool the water, which saves on energy consumption.

Because wastewater treatment operations take much space, they are located outdoors and this implies that the system must be able to operate at seasonally varying temperatures. The advantage is low energy consumption, but the disadvantage is low productivity during colder months.

***Integrated Cultivation Systems Summary***

Integrated systems have a variety of applications and can be scaled with relative ease (from small aquariums to large bodies of water) compared with other cultivation systems. The environmental barriers to commercial scalability of integrated cultivation facilities include the potential need for appropriate land adjacent to polluted waterbodies, likelihood of requiring biomass transport to downstream processing facilities, life cycle of pond liners (where used), and potential impact on aquatic life in the immediate vicinity of the facility.

**Pathways for Biomass Harvesting**

Once an algal culture reaches maturity, the biomass is harvested from the culture medium and dried in preparation for conversion. Biomass harvesting may be one of the more contaminating processes in the production of algae-based biofuels. At this stage, algal biomass from the preceding cultivation system typically carries a high water content and, in most cases, is not suited for conversion to biofuel products until it has undergone some degree of dewatering and drying.

There are three systemic components of the harvesting process: biomass recovery, dewatering, and drying. Some pathways employ all three processes, whereas others may only employ one or two of these processes.

**BIOMASS RECOVERY**

Recovering the algae and disposing or recycling of the process water represent two energy intensive and potentially significant environmental challenges to sustainable algae-based biofuel production.

***Recovery Techniques and Characteristics***

There are several techniques for recovering algal biomass, the implementation of which may vary depending on existing pond conditions or PBR design. The most commonly implemented techniques are flocculation, dissolved air flotation, centrifugation, microfiltration, and decantation, each of which is discussed briefly. Additional techniques—discrete sedimentation, membrane filtration, phototactic autoconcentration, tilapia-enhanced sedimentation, tube settling, and ultrasonic separation—may also be considered viable pathways to biomass recovery, but are beyond the scope of this report.

**Flocculation**

Flocculation is a process, often implemented with the help of flocculating agents or flocculants (chemicals of natural or synthetic origin), that causes the coagulation of algal cells into small clumps, known as flocs, allowing for sedimentation and easy extraction from the culture medium.

Flocculation is the historically preferred recovery technique for its simplicity and variety of mechanisms, including autoflocculation, bioflocculation, electroflocculation, foam flocculation, inorganic chemical flocculation, ozone flocculation, and polyelectrolyte flocculation. These and other mechanisms of flocculation are familiar to the engineering of waste and water treatment. Bioflocculation, chemical flocculation, and electroflocculation are discussed here to introduce the range of mechanisms and the potential environmental impact of flocculation in general.

*Bioflocculation*—an approach to water treatment that can be traced back 2,000 years.<sup>93</sup> It uses naturally occurring, biodegradable polymeric (e.g., Chitosan, sodium alginate) or a microbial (e.g., Pestan) flocculants to coagulate the algal cells.

*Chemical flocculation*—uses a chemical additive, such as aluminum sulphate or cationic polyelectrolytes, to initiate flocculation (see Pathway Map A-II).<sup>94</sup> Chemical additives have varying abilities to enhance flocculation rate, which may depend on the effects of temperature, nutrient conditions, and pH of the culture medium. Polyelectrolyte flocculation, widely applied since the 1960s to waste and water treatment, is favored in the industry for its efficiency and for the water solubility of the synthetic organic polymeric flocculants.<sup>95</sup>

*Electroflocculation*—uses electricity and metal ion flocculants to coagulate the algae cells. Culture mixing and higher electrical input currents

improve removal efficiencies in shorter periods of time.<sup>96</sup>

Serious drawbacks to the application of chemical or synthetic flocculants include the contamination of process wastewater and residual biomass.<sup>97</sup> Although synthetic flocculants are often more efficient than naturally occurring flocculants, their long-term effects on human and environmental health are still unknown.<sup>98</sup> Algal biomass extracted with the help of chemical flocculants may carry high levels of associated toxins. Chemical flocculants aluminum sulphate and cationic polyelectrolytes contain high concentrations of aluminum and polyacrylamide residues, respectively. These can be toxic to animals and will affect the culture medium in such a way that



**Figure 16:** Algae and phosphorous after undergoing dissolved froth flotation. Phosphorus from the effluent of wastewater treatment ponds can be removed by chemical precipitation of the soluble phosphorous and then the solids, formed together with the algae, are separated from the water by flotation.

Source: [www.armatec.co.nz/products/water.asp](http://www.armatec.co.nz/products/water.asp)

it must first be treated before being released or recycled through to the cultivation system.<sup>99</sup>

The use of transgenic algae that have been modified to autoflocculate under predetermined culture conditions may also be a solution for lowering barriers to efficient and sustainable biomass recovery.<sup>100</sup> In this context, the application of transgenic algae may need to be restricted to closed PBRs.

Understanding and controlling the mechanisms of bioflocculation may help improve efficiency of biomass recovery without the associated impacts of toxic chemicals.<sup>101</sup> Though perhaps less efficient, flocculation by pH adjustment, without a chemical flocculant or subsequent alteration of the culture medium, could be a safer way to recover algal biomass.<sup>102</sup> Another consideration is that all flocculation mechanisms require some level of pH regulation; therefore, under the right conditions, pH adjustment or bioflocculation could be the more sustainable mechanisms.

#### **Dissolved Air Flotation**

Dissolved air flotation (also known as froth flotation, foam separation, or foam flocculation) is an established technology for sewage sludge thickening. In the context of algal biomass recovery, dissolved air flotation uses a flocculant and the aid of pressurized air bubbles (heat or entrained air) to force the algal cells to cluster and float to the water surface (Figure 16) where they can be removed by a skimming device.<sup>103</sup>

Dissolved air flotation is commonly practiced in the municipal wastewater and paper industries. Both industries process very large volumes of water for removal of solids and do so economically using dissolved air flotation systems, indicating the potential viability of this method in commercial-scale biomass recovery from algae cultivation systems.<sup>104</sup> The environmental impact of this process includes the energy needed for pressurization and the type of additives used for adjusting the pH and decreasing the surface tension of the culture medium (which allows the cells to cluster and float to the surface).<sup>105</sup>

#### **Centrifugation**

A centrifuge, generally driven by a motor, puts the algal culture in rotation (applying force) to evenly distribute the water and biomass by greater and lesser density. Centrifugation is typically a high-energy process considered impractical for large-scale harvesting; however, large industrial centrifuges are commonly used in water and wastewater treatment to dry sludge.

After centrifugation, algae on the surface can be removed by decantation, and the remaining culture medium, known as centrate, can be recycled through the cultivation process without pretreatment because no chemicals are used in this process. However, the high energy required for operation could be prohibitive if applied as a primary dewatering technique for commercial-scale biomass harvesting. An alternative application could be to use centrifugation as a secondary or tertiary method of dewatering.

#### **Microfiltration**

Microfiltration, or microscreening, is a basic approach to biomass recovery whereby algal cells are filtered through microscreens to be separated from the growth culture. Conventional microscreens could prove impractical because of the small size of microalgae (typically 1–30 microns; e.g., *Chlorella* is 2–10 microns) and the high rate at which the algae would obscure the filter media.

Vibrating microscreens are rotating, backwashed, fine-mesh screens with the ability to harvest continuously and by cell size (i.e., maturity). This helps to maximize yields because immature algae are passed through, leaving mature, oil-rich algae to be harvested (see Pathway Map B-II).<sup>106</sup>

Microfiltration does not necessarily employ chemicals and does not require treatment of filtered water before it is recycled to the cultivation system, along with the immature, unharvested algal cells.

#### **Decantation and Vacuuming**

Decantation is the draining of the culture medium to allow the algal cells to settle. The settled biomass can be harvested by vacuum, a process meant to be gentle on the algal cells (see Pathway Map E-II). In



most cases, decantation and vacuuming are low-impact, secondary recovery processes implemented after flocculation, centrifugation, or dissolved air flotation.

### ***Environmental Impacts of Biomass Recovery***

Environmental issues concerning the scalability of biomass recovery include the toxicity of chemical additives and output water quality, and are discussed here in terms of water and energy.<sup>107</sup>

#### **Water**

Ideally, recovered process wastewater is recycled back into the cultivation system or released downstream at a quality equal to or greater than before it entered the system. Some recovery methods may allow for water recycling or release without treatment. Although chemical flocculation is certain to contaminate the water, the quality of process water from dissolved flotation and certain flocculation mechanisms will depend on the type of additive used.

Managing the presence of chemical and other contaminants in the recovery process will be key to producing high-quality biomass with the least environmental impact. The greater the presence of metals or chemicals, the more refining needed to prepare the fuel for market consumption and the greater the output of contaminated water or unwanted byproducts such as acids or soap.<sup>108</sup>

The use of metal catalysts or chemical solvents in harvesting processes will likely require treatment of process wastewater before reuse or release. Proper wastewater management will limit any environmental pollution from the release of wastewater into a receiving waterbody. Otherwise an algal cultivation system may utilize and treat wastewater only for the water to be recontaminated (on a smaller yet still relevant scale) by harvesting and conversion processes that use chemical or metal additives.

Additionally, the presence of heavy metals is known to inhibit catalyst function in subsequent conversion processes.<sup>109</sup> In many cases, there seem to be natural alternatives to chemical or metal catalysts. Adjusting pH is a toxin-free alternative to chemical flocculation. Therefore,

employing biomass recovery techniques that minimize or eliminate the use of chemical and metal flocculants, catalysts, or solvents will improve biomass quality and reduce the need for wastewater treatment.

#### **Energy**

The energy requirement for biomass harvesting is a function of several issues, including the type of algae; its harvest density; and the harvest techniques for biomass recovery, dewatering, and drying. The energy and maintenance needed for biomass recovery could impose obstacles to sustainable scalability for certain recovery methods, specifically with respect to the management of process water exposed to chemical additives. However, availability of reliable data is limited; further research and analysis are needed to determine the true impact of biomass recovery.

#### **DEWATERING**

With the exception of heterotrophically cultivated biomass, algal biomass typically has a high water content ( $\leq 99$  percent) and, in most cases, is not suited for conversion to biofuel products until it has undergone some degree of dewatering. Dewatering decreases the moisture content of the biomass by draining or mechanical means and can be implemented either before recovery (e.g., integrated systems) or after recovery (e.g., closed PBRs). The need for dewatering is largely dependent upon the desired moisture content and is therefore often paired with other harvesting processes or, in some cases, is excluded altogether.

#### ***Dewatering Techniques and Characteristics***

The dewatering process can increase biomass solids content up to approximately 20 percent via a draining tank or screw press. Recovered biomass can be directed to a vessel, such as a stainless steel tank, where the water settles to the bottom and is drained out of the tank.

A mechanical screw press expels water with pressure and, like the draining tank, directs wastewater to a treatment facility if necessary and then back to the cultivation facility. Dewatering uses few inputs, if any, which are restricted to the energy required for operating and maintaining the screw press or draining tank.



### ***Environmental Impacts of Dewatering***

Environmental issues concerning the scalability of dewatering are discussed in terms of water and energy. As dewatering involves the expulsion of large quantities of water that possibly contain chemicals or algae particles (native, exotic, or transgenic), its management could have a negative effect on the environment.

#### **Water**

Some facilities could potentially develop the capacity to fully reuse or release process wastewater—where no exotic or transgenic algae were used for cultivation and no chemicals were used in the recovery process—while others may need to administer treatment of wastewater for reentry into public waterways or the natural environment. A very high percentage of process water can be recovered and recycled back through the cultivation process; however, the applicability of full water recovery at commercial scale is still to be determined.

#### **Energy**

Reduced water content increases the energy density of the biomass; therefore, dewatering is conducted primarily to minimize the amount of thermal energy needed for drying. Whereas drier biomass may be advantageous, dewatering plus drying should be weighed against the maintenance and energy demands of oil extraction and conversion technologies that bypass both processes by accommodating biomass with high moisture content.

### **DRYING**

As mentioned, the level of drying achieved depends on the requirements of subsequent processes. Some conversion methods require a high solid content ratio, while others do not. These differences will be touched upon later in the report.

### ***Drying Techniques and Characteristics***

There are several ways to dry algal biomass. Some techniques are well understood while others are still undergoing development. Solar, drum, freeze, spray, and rotary drying are among some of the approaches being tested with algal biomass. The brief descriptions below provide a general understanding of the variation of these techniques.

### **Solar Drying**

Solar drying of algae is a basic process where the water flow is turned off to allow algae to drain and dry naturally by the sun. Drying time can be reduced with the help of fans. Biomass from an algal turf scrubber system (see Pathway Map E-I), for example, can be harvested with 1–3 hours of solar drying and vacuum recovery.

### **Drum Drying**

Drum dryers are a more technologically advanced approach that expedites biomass drying, whereby the dewatered algae is sent over a series of heated drums (see Pathway A.II). This drying process thins out the algal biomass, creating a product that might resemble an algae paper, which could be rolled up for easy storage and shipment. According to Ron Putt (2007:13) of Auburn University, the biomass must be dried to a point of at least 90 percent solids content in order to avoid spoiling during storage and shipment.

The latent heat required to increase solids content from 20 percent to 90 percent is about 170 kWh/acre per day. Gas-fired forced air could be used for heating the drum dryers by way of a diesel engine or generator discharging thermal power.

### **Freeze Drying**

Freeze drying, or lyophilization, removes water content from the biomass under a low air pressure vacuum, causing water inside the cells to slowly vaporize. The process ends in a condenser, with the algae solidly frozen, maintaining cell structure without degradation.<sup>110</sup>

### **Spray Drying**

Spray drying, which can be applied to pond-cultivated biomass, is the operation most ideally suited for harvesting dry algae in the powder or granular form for use in dietary supplements.

### **Rotary Drying**

Steam tube rotary dryers are convective-type dryers that allow steam (heated by an external source) to rise as it is replaced by cool air, creating a current of circulating hot air. Rotary dryers are sometimes

## Summary of Environmental Issues Related to Biomass Harvesting

There are several environmental benefits, concerns, and unknowns with which sustainable biomass harvesting will likely be challenged, many of which can already be identified. The criteria for sustainable biomass harvesting should consider potential environmental toxicity of chemical additives, management of output water, and implications of energy-intensive drying techniques.

### ENVIRONMENTAL BENEFITS

- ▶ A very high percentage of water can be recovered and recycled through to the cultivation process
- ▶ Process wastewater released downstream could potentially be at a quality equal to or greater than before it entered the cultivation process

### ENVIRONMENTAL CONCERNS

- ▶ Harvested biomass may require storage and transport for downstream processing
- ▶ Quality or toxicity of process water will depend on the type and quantity of additives used and will determine need for treatment of process wastewater before reuse or release
- ▶ Presence of exotic or genetically modified algae in process wastewater could threaten native ecosystems

- ▶ Biomass moisture content may affect energy and chemical inputs in downstream processing
- ▶ Energy-intensive processes may negatively impact the energy balance
- ▶ Identified environmental impacts might be accepted as trade-offs for more efficient harvesting mechanisms

### ENVIRONMENTAL UNKNOWNNS

- ▶ Rate (percent) of water recovery
- ▶ Rate (percent) of chemical recovery
- ▶ Facility scalability
- ▶ True impact of energy-intensive drying systems
- ▶ Current practices for water management: Are process outputs recycled or treated and released?
- ▶ Whether research and development should target improving efficiency of drying methods or improving the capacity of conversion technologies to accommodate wet biomass
- ▶ Potential benefits or concerns associated with emerging academic or nonconventional harvesting pathways

paired with fermentation processes, such as heterotrophic cultivation (see Pathway Map D-II).

### *Environmental Impacts of Drying*

Environmental issues concerning the scalability of the drying process are centered on water expulsion and energy consumption. The impact of water expulsion may also be of some consequence, though not discussed here for lack of data. Beyond water and energy, environmental impacts are likely to be minimal.

#### **Water**

Drying technologies either evaporate or collect by vaporization the remaining water content. Any potential effect this might have on water or air quality will depend upon the process used and should be considered in a feasibility study.

#### **Energy**

Drying practices of today appear to favor drum dryers over solar or freeze drying; however, as drum dryers require considerable energy inputs, rotary drying and other emerging methods may soon outperform conventional ones. The development of a biomass conversion process that accommodates feedstocks with high moisture content also has the potential to eliminate the need for these drying methods, possibly leading to a significant reduction in process energy consumption.

Although the environmental impacts of biomass harvesting appear to be confined mostly to water and energy management, the question remains as to whether research and development should be targeted to improve the efficiency of drying

methods or the capacity of conversion technologies to accommodate wet biomass.

### Pathways for Algal Oil Extraction

In recent years, many calculations for theoretical oil yields from algae have been made. The biomass generated from algal growth and cell division is known to reach oil contents of up to 80 percent, making algal biomass an appealing candidate for biofuel feedstock.<sup>111</sup> Although the actual oil content (2–80 percent), measured in gallons/acre/year, will depend on many parameters, there is certainly a scientific basis allowing projection of potential yields that are orders of magnitude larger than current biofuel technologies.<sup>112</sup> However, while there is a range of practical and theoretical oil yield estimates publicly available, in this section we identify the technologies and environmental impacts associated with extracting oil from the algal cells, and oil yields will only be referred to in terms of technological efficiencies.

At this stage in the biofuel production process, the percent yield of total available oil from the biomass will depend on the efficiency of the extraction method used. In some instances, technologies may be favored for their superior performance (e.g., chemical extraction) over less efficient technologies (e.g., mechanical extraction), despite higher environmental costs.

Depending on the desired fuel product, algae will undergo a variety of treatments to manipulate the oil and residue. This section discusses methods for extracting algal oil for conversion to biodiesel. Algal oil is typically extracted by mechanical, chemical, or electrical means, whereas bio-oil is extracted with the use of chemicals and high temperatures. This difference is important because each pretreatment yields either algal oil or bio-oil, two chemically distinct feedstocks—possibly with differing environmental implications—which are subsequently converted to different fuel products. The next section will discuss oil and residue conversion pathways, including pretreatments for bio-oil.

#### *Extraction Techniques and Characteristics*

Oil extraction from algal biomass yields algal oil (triglycerides or TAG lipids) and residue (carbohydrates, proteins, nutrients, ash). Algal oil extraction can be achieved via a number of techniques such as mechanical expulsion, solvent extraction, or supercritical fluid extraction. Osmotic shock and sonication are less common methods and are only discussed briefly.

#### **Mechanical Expulsion**

Mechanical technologies for extracting algal oil include the screw press, extruder and expander, and pulverization in a mortar. In the mechanical expulsion process, oil is expelled from dried algal cells by one or more of these methods (see

<b>Supercritical Fluid</b>	<b>Critical Point (Liquefaction Temperature °C)</b>	<b>Atmospheric Pressure (atm)</b>
Water	374 degrees	218 atm
Carbon dioxide	31 degrees	73 atm
Propane	97 degrees	42 atm
Ethanol	241 degrees	61 atm
Methanol	240 degrees	80 atm
Ammonia	133 degrees	111 atm

Data Source: [http://www.econ.iastate.edu/outreach/agriculture/programs/2001\\_Renewable\\_Energy\\_Symposium/Aurand.pdf](http://www.econ.iastate.edu/outreach/agriculture/programs/2001_Renewable_Energy_Symposium/Aurand.pdf)

Pathway Map A-III). Machines that combine these technologies for increased extraction efficiencies are also available. Process inputs are basic electricity to power the machinery.

Possible innovations could help overcome the inefficiencies of mechanical technologies, including genetically modifying algal strains to have weaker cell walls that can be broken under lower pressures or low-heat pretreatment.<sup>113</sup>

A combination of mechanical expulsion and chemical solvents also holds the potential to increase efficacy of the extraction process. Breaking the cells under the high pressure of a mechanical press may cause the fusion of lipid droplets with cellular membrane material that leads to a loss of oil. When mechanical methods are paired with hexane solvent extraction, this “lost” oil can be recovered.<sup>114</sup>

#### **Solvent Extraction**

Hexane (or chloroform) is a relatively inexpensive chemical commonly used in oil extraction from soybeans and other plants and is now being explored for its efficiency in expelling oil from algal cells. Hexane solvent extraction mixes hexane with the algal biomass. The oil dissolves in the hexane and the biomass can be filtered out from the medium through distillation (see Pathway Map D-III). Although this process can be used effectively in isolation, coupled with press expulsion these two processes are capable of extracting most of the total available algal oil.

#### **Supercritical Fluid Extraction**

In the supercritical fluid extraction process, oil is extracted from the algal cells with a solvent, such as methanol or liquefied CO<sub>2</sub>, and heated under pressure up to or above its critical point (see Pathway Map E-III). The process has the efficiency and ability to isolate oil components leading to the extraction of almost 100 percent of the oils.<sup>115</sup>

The liquefaction of a solvent for supercritical extraction is often an energy-intensive process. The temperature and pressure (critical point) at which the fluid liquefies vary depending on the type of solvent used (Table 3), which would determine respective energy inputs.

#### **Osmotic Shock**

Osmotic shock is the sudden reduction in the movement or concentration of water across the algal cell membrane. The stress from the rapid change in movement, created by the addition of high concentrations of a solute or other additive (e.g., salt, substrates, neutral polymers such as polyethylene glycol, dextran) causes the cells to rupture, releasing the oil.

#### **Sonication**

Sonication uses an ultrasonic reactor (sonicator) to make acoustic shock waves and liquid jets that induce algal cell walls to break and release their contents into the medium without the use of toxic solvents (see Pathway Map C-III).<sup>116</sup>

#### ***Environmental Impacts of Extraction***

Environmental issues concerning sustainable oil extraction include recalcitrant biomass residue, chemical solvents, and energy demand.

#### **Recalcitrant Biomass Residue**

After the main portion of the biomass is separated into oil, protein, and starch, there will be a portion left over consisting mostly of metals, salts, lignin, and other recalcitrant matter that will need to be managed. The environmental implications of the processing, such as via anaerobic digestion or disposal of recalcitrant biomass, will need to be considered for commercial-scale systems.<sup>117</sup>

#### **Chemicals**

Because of the inefficiencies of mechanical expulsion technologies (where up to 10–25 percent of the oil can be lost), stronger consideration may sometimes be given to chemical extractions, such as supercritical fluid extraction, to provide an improved yield.

The specific solvent used (e.g., CO<sub>2</sub>, methanol, hexane), if any, will determine the environmental impact of the extraction process.<sup>118</sup> Since volatile chemical solvents have inherent health and safety problems as well as environmental toxicity, the feasibility of storage, handling, and disposal may have additional implications.<sup>119</sup> For example, the hexane extraction process can cause lethal explosions in laboratory and commercial settings.

## Summary of Environmental Issues Related to Algal Oil Extraction

There are a few environmental issues with which sustainable algal oil extraction will likely be challenged. Known environmental benefits and concerns are summarized below. At a minimum, the criteria for sustainable oil extraction should consider energy inputs and potential environmental toxicity and safety concerns of chemical solvents. Because of the limited availability of information on chemical and energy inputs to algal oil extraction techniques, some impacts may have been unintentionally overlooked in this summary.

### ENVIRONMENTAL BENEFITS

- ▶ Impacts may be relatively easy to identify and avoid

### ENVIRONMENTAL CONCERNS

- ▶ Most efficient extraction methods require biomass with high solids content, meaning more energy required for biomass drying
- ▶ Chemical solvents have inherent health and safety implications
- ▶ Environmental toxicity of chemical solvents will restrict storage, handling, and disposal methods
- ▶ The specific solvent used will determine the environmental impact of an extraction process
- ▶ Recalcitrant biomass residue, which will likely increase proportionately as scale increases, will need proper management and possibly disposal

### ENVIRONMENTAL UNKNOWNNS

- ▶ Energy demand at scale, including indirect energy inputs

Therefore, the environmental impacts of hexane and other chemicals used at commercial scale will need to be better understood to ensure a safe and sustainable extraction process.

### Energy

Primary energy inputs of environmental relevance are those of heat, electricity, and pressure, which will vary depending on extraction technique, as well as the liquefaction of CO<sub>2</sub> and other solvents intended for supercritical fluid extraction.

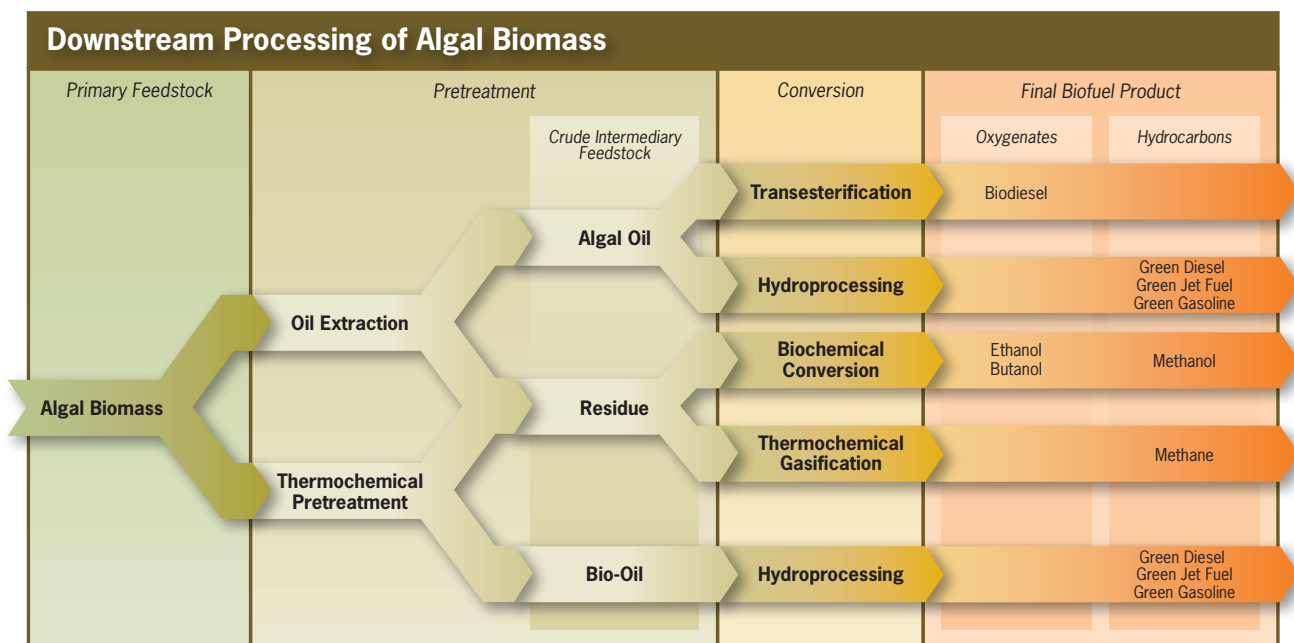
Whereas algal oil extraction draws largely from existing technologies, significant technical and process engineering challenges may need to be solved before a truly scalable and safe means of oil extraction is established.<sup>120</sup> Super Mechanical methods are perhaps the more mature of oil extraction techniques; however, they are not the most efficient and thus not particularly suited to commercial-scale practices. Other processes that require special equipment for containment and pressure could increase energy demand at scaleup.<sup>121</sup> The environmental costs associated with various extraction techniques need to be measured to determine the sustainability of commercial-scale extraction.

### Pathways for Oil and Residue Conversion to Biofuels

Once the biomass is separated into raw algal oil and residue, the energy content of the two components can be thermally or biologically transformed to liquid or gaseous fuels or solid coproducts. Conversion pathways include transesterification, fermentation, anaerobic digestion, gasification, pyrolysis, liquefaction, and hydroprocessing. These pathways are discussed below according to whether the technology is biochemical or thermochemical. Industry may categorize these technologies in a different manner; however, the goal of this section is not so much to define pathways (e.g., pretreatment vs. conversion) as to identify major environmental issues associated with them.

These conversion pathways are nearly identical to those for converting first- and second-generation biofuel feedstocks; hence, this section does not focus on differentiating the conversion of algal biomass from other feedstocks.<sup>122</sup> The descriptions are kept brief—the nuances of each conversion system are generally excluded—while greater emphasis is put on environmental benefits, concerns, and unknowns in the event such a technique is applied to an algae biofuel production pathway. These descriptions are meant to





**Figure 17:** Pathways for the downstream processing of biomass can follow a number of routes. The chemical makeup and quality of intermediate products (residue, algal oil, bio-oil) will play a role in determining which processes may be used, as well as how great the environmental externalities may be of a specific pathway.

provide the reader only with a general understanding of the range of processes being explored by algae biofuel producers and the relationship between the conversion process and preceding (upstream) processes in a production pathway. An overview of potential bioproducts is also presented to provide context for this discussion about oil and residue conversion pathways.

### Conversion Pathways

Depending on the condition or quality of the biomass and the intended fuel application, a range of conversion pathways could be pursued. Bio-oil is chemically distinct from algal oil and must therefore be converted to biofuel under different conditions. When biomass is pretreated thermochemically, via pyrolysis or liquefaction, it produces intermediate products bio-oil and residue.

Once the biomass is separated into oil and residue (nutrients, proteins, carbohydrates, ash), transesterification can convert algal oil to biodiesel; hydroprocessing can convert algal oil and bio-oil to green or renewable biofuels; and much of the residue can be biochemically or thermochemically converted to a gaseous fuel or a solid, nutrient-rich bioproduct (Figure 17).

### Biofuel Products

Conversion processes are of varying efficiency—depending on reaction temperature, pressure, heating rate, and catalyst type, as well as algal species and quality of biomass—theoretically converting algal biomass (or components of biomass) into several possible biofuels and coproducts. There are essentially two categories of biofuels being produced today: hydrocarbons and oxygenates.

### Hydrocarbon Biofuels

Hydrocarbons are fuels such as gasoline, diesel, and jet fuel that do not contain oxygen. Bio-derived hydrocarbon fuels are products of thermochemically converted algal oil or bio-oil and are sometimes referred to as green or renewable gasoline, diesel, and jet fuels. Fuel properties (Table 4) will differ based on biomass origin, fuel type, and country specifications. One of the foremost qualities of hydrocarbon biofuels is that they are drop-in replacements for existing petroleum-based transportation fuels. Other hydrocarbon fuels include methane ( $\text{CH}_4$ ), ethane ( $\text{C}_2\text{H}_6$ ), and propane ( $\text{C}_3\text{H}_8$ ).

Table 4. Comparison of Renewable Properties							
Fuel Properties*	Oxygenate Biofuels			Fossil Fuel	Hydrocarbon Biofuels		
	Butanol	Ethanol	FAME Biodiesel	Petroleum Diesel	Green Diesel	Green Gasoline	Green Jet Fuel†
Boiling point (°C)	117	78	182 to 338		180 to 380	35 to 215	150 to 300
Flash point (°C)	29 to 37	130	125 (100 to 170)	55 <sup>b</sup> 60 to 80	65		38
Cloud point <sup>a</sup> (°C)			-3 to +12 (-5 to +15)	-5 (-15 to +5)	-20 to -10 (-30 to +10)		
Pour point (°C)			-15 to +10 (-15 to +16)	-35 to -15	-15 to 0		
Freeze point (°C)	‡	-117 to -114	0 (32°F)		-8		-47 <sup>c</sup>
Embodied energy (MJ/L)			37.27		44		42.8
Drop-in replacement	No	No	Yes	–	Yes	Yes	Yes

\* Temperatures are feedstock dependent. These numbers do not necessarily reflect algal biomass as the feedstock.

<sup>a</sup> The cloud point, or gel temperature, depending on what oil was used as a feedstock.

<sup>b</sup> Specification for U.S.

† Source: UOP A Honeywell Company

<sup>c</sup> SPK Jet A-1

‡ Butanol has a melting point of -90°C

Table 5a: Transesterification, Bioproducts, and Applications			
Pathway Process	Feedstock	Chemical Product*	Application
Transesterification	algal oil	fatty acid methyl ester (FAME) biodiesel	diesel engines
		glycerol, fatty acid ethyl ester (FAEE)	glycerol, possibly to soap or feedstock

\* For detailed list of bio-based products, see table in Wang et al. (2007:1203) or Kamm and Kamm (2007:185).

### Oxygenate Biofuels

Bio-derived oxygenates are fuels that are typically made either via transesterification of algal oil, yielding biodiesel (or FAME biodiesel), or biochemical fermentation of biomass sugars and starches, yielding biobutanol and bioethanol. Alcohol-based biofuels (e.g., bioethanol) are not directly compatible with existing infrastructure because they corrode pipelines and storage infrastructure and are not amenable to blending with FAME biodiesel or petroleum diesel.

The conversion pathways for producing hydrocarbon and oxygenate biofuels will differ, as will their environmental impact.

### Other Bioproducts

In addition to fuel products, most conversion processes yield products of higher, lesser, or no value (Tables 5a–5d) that have varying environmental significance. The most common of these products are discussed below in terms of their usability and potential for impacting the environment under commercial-scale biofuel production.

Coproducts refer to process outputs of low or high value (e.g., crude oil, biofuel, glycerol, biopolymers) that have an existing or emerging application (market).<sup>123</sup> Byproducts refer to process outputs with little or no value (existing market) that are often toxic and must be disposed of (e.g., CO<sub>2</sub>, tar, certain acids),

or for which pretreatment is required in order to reclaim value or viable reuse (e.g., wastewater).

### TRANSESTERIFICATION

Transesterification converts raw algal oil to biodiesel for direct consumption by unmodified diesel engines (see Pathway Map A-IV).<sup>124</sup> There is nothing unique about the transesterification of algal oil compared with that of conventional vegetable oils.

#### *Conversion Characteristics*

Oils can be converted via acid-catalyzed, alkali- or base-catalyzed, or enzymatic transesterification.<sup>125</sup> An acid catalyst, such as sulfuric acid, is used when the oil has high acid value. When the oil has low acid value, an alkali catalyst may prove more effective.<sup>126</sup> Transesterification can be performed continuously or using a batch process.

The main byproducts of transesterification are fatty acid ethyl ester (FAEE) and glycerol (also known as glycerin or glycerine) (Table 5a). During conversion, glycerol is periodically or continuously removed from the reaction solution in order to drive the equilibrium reaction toward completion.

The presence of methanol, the cosolvent that keeps glycerol and soap suspended in the oil, is known to cause engine failure. To prevent this, centrifugation washes biofuels from the soap (and glycerol). Dry bubbling, a longer process (2–3 days) that promotes the evaporation of methanol, can expedite byproduct separation and settling, ridding the biofuel of soap.

#### *Environmental Impacts of Transesterification*

Although alkali-catalyzed transesterification achieves higher yields at higher reaction rates than acid catalysts, the reaction also requires a feedstock with minimal impurities (such as moisture and free fatty acids) and leads to soap formation (a process caused by saponification).<sup>127</sup> Higher quality feedstocks are often environmentally costly to prepare (e.g., algae cultivation uses nonpolluted water; harvested biomass is dried to a higher percent solids content). Acid-catalyzed and enzymatic transesterification accommodate feedstock with a greater degree of impurities and do not cause the formation of soap.

Glycerol could be a viable feedstock for algae cultivation by heterotrophic fermentation; however, glycerol currently has only a niche market. The output

quantity of glycerol is also relative to that of biodiesel, meaning that the more biodiesel produced via transesterification, the more glycerol produced. Thus, the current market for glycerol would not likely be able to accommodate commercial-scale biodiesel production via transesterification. Without an alternative market, commercial-scale production of algae-based biodiesel may lead to excess glycerol that will need to be managed, stored, and disposed.

### BIOCHEMICAL FERMENTATION

Biochemical conversion breaks down sugars in the residue using enzymatic or chemical processes such as fermentation or anaerobic digestion. Fermentation is the more common biochemical approach to algal biomass conversion. Primary fuel products of biochemical conversion include methanol, ethanol, butanol, and hydrogen.

#### *Conversion Processes and Characteristics*

Biochemical conversion technologies, as described by NREL, involve three basic steps: 1) converting biomass to sugar fermentation feedstock; 2) fermenting the feedstock using biocatalysts (microorganisms including yeast and bacteria); and 3) processing the fermentation product to yield fuels, chemicals, heat, or electricity.<sup>128</sup>

#### **Anaerobic Digestion**

Anaerobic digestion is a biochemical process that converts residue to hydrogen for biofuel. Common coproducts are methane and a nutrient-rich fertilizer or feedstock. Anaerobic digestion is not currently a popular pathway for algal residue conversion; however, it could be an option for the processing of recalcitrant biomass residue from other conversion processes.

#### **Fermentation**

Fermentation can be defined as biochemical decomposition in the absence of air, essentially converting sugar to carbon dioxide and alcohol. Anaerobic fermentation (without oxygen) can transform the algal residue into butanol and methanol, whereas aerobic fermentation yields hydrogen and ethanol. Anaerobic conditions allow for sugars to be broken down in just one step, while aerobic conditions tend to require additional

Table 5b: Biochemical Conversion Processes, Bioproducts, and Applications			
Pathway Process	Feedstock	Chemical Product*	Application
Anaerobic digestion	residue	hydrogen	biofuel
		methane (CH <sub>4</sub> ) liquid fertilizer	medium Btu gas for electricity and heat for recycling as eutrophied process water
Anaerobic fermentation	residue	hydrogen, ethanol	biofuel (ethanol is suitable for light vehicles)
		CO <sub>2</sub> , methane (CH <sub>4</sub> ) acetone, acetic acid, lactic acid, other acids	reuse in production processes
Aerobic fermentation	residue	butanol, methanol	biofuel
		CO <sub>2</sub> , methane (CH <sub>4</sub> ) acids, industrial alcohol	reuse in production processes
		potable alcohol	bottled potable alcohol

\* For detailed list of bio-based products, see table in Wang et al. (2007:1203) or Kamm and Kamm (2007:185).

steps (citric acid or Krebs cycle and electron transport) to complete the conversion process.

Byproducts of the fermentation process are CO<sub>2</sub>, methane, water, and several acids, including acetic and lactic acids (Table 5b). Anaerobic fermentation also yields acetone. Methane is suitable for electricity and heat, whereas other liquid byproducts, such as acetone, are suitable for recycling as eutrophied process water.<sup>129</sup>

### ***Environmental Impacts of Biochemical Fermentation***

It is not clear whether the sugars required for fermentation would be environmentally burdensome or financially inhibitive at commercial scale. This may depend on their origin (e.g., irrigated terrestrial crops) and what preparatory processes are employed to ready the sugars for fermentation.

In addition, the organic byproducts (e.g., lactic acid) are toxic, requiring appropriate storage and disposal, and the potential GHG implications of released methane and carbon dioxide could also be a concern.

### **THERMOCHEMICAL CONVERSION**

Endothermochemical conversion involves the consumption of energy to convert a fuel source (i.e., biomass) into a different chemical state (i.e., oil and residue). Exothermochemical conversion (releasing energy) via combustion to generate power is not

within the scope of this report. The characteristics of endothermochemical conversion processes are discussed along with associated environmental issues.

### ***Conversion Processes and Characteristics***

Pyrolysis and liquefaction pretreat algal biomass to yield intermediary fuel products bio-oil and residue (as opposed to algal oil and residue from the algal oil extraction process). The residue can be either biochemically converted (e.g., fermentation) or thermochemically converted via gasification; the bio-oil can be upgraded by hydroprocessing.

### **Gasification**

Gasification is a thermochemical process that, in the near absence of oxygen, converts organic material into a combustible gas called producer or synthesis gas (syngas) (see Pathway Map A-IV). Syngas, comprised of mainly of CO, CO<sub>2</sub>, H<sub>2</sub>, water and tar vapors, and ash particles, contains 70–80 percent of the energy originally present in the biomass feedstock.<sup>130</sup> With proprietary catalysts, syngas yields fuel gases H<sub>2</sub>, ethanol, methanol, and dimethyl ether (DME).<sup>131</sup>

Conventional catalytic gasification occurs at temperatures 800–1,000°C or higher,<sup>132</sup> whereas supercritical water (hydrothermal) gasification occurs at 347°C with a metal catalyst or at

697°C with a carbonaceous or alkali catalyst.<sup>133</sup> This is important to note because lower reaction temperatures lead to smaller reactors and lower energy inputs. Therefore, the difference in energy consumption between the two approaches to gasification could be substantial.

Conventional gasification also requires dry biomass with moisture content no higher than 15–20 percent. Current efforts to perform supercritical water gasification with wet biomass could significantly reduce environmental impacts as such a development would bypass energy-intensive dewatering and drying processes and reduce the need for high-temperature reactors.<sup>134</sup>

The makeup of syngas (the primary product of gasification) tends to vary based on the type of feedstock, moisture content, type of gasifier used, gasification agent, and the temperature and pressure in the gasifier.<sup>135</sup> Typical gaseous components, such as CO, CO<sub>2</sub>, H<sub>2</sub>, CH<sub>4</sub>, and N<sub>2</sub>, can be used in turbines and boilers or as feed gas for the production of liquid alkanes by Fischer-Tropsch (FT) synthesis.

According to the DOE Office of Energy Efficiency and Renewable Energy (EERE), as of 2004 there had not been much emphasis on understanding the potential impact of the byproducts of gasification, particularly of tar, nitrogen (N), sulfur (S), chlorine (Cl), and alkali species, on downstream unit operation, and final product quality.<sup>136</sup>

### Pyrolysis

Pyrolysis, also known as thermal cracking, is a thermochemical pretreatment process of induced decomposition (see Pathway Map B-IV), conducted in the relative absence of oxygen. This process is applied at temperatures above 430°C (800°F); the typical range is between 500–600°C, at 0.1–0.5 MPa (megapascal) of pressure. When the off-gases are cooled, liquids condense, producing oil and contaminated water. This organic oil is often referred to as pyrolysis oil or bio-oil.

Slow pyrolysis produces a black, tarry oil residue, while fast pyrolysis outputs dark-brown, low-viscosity (and therefore higher quality) oil.<sup>137</sup> Data reporting yields for fast pyrolysis vary extensively from 18–80 percent efficiency.<sup>138</sup>

Carbon monoxide, charcoal, phenol-formaldehyde resins, and wastewater are common byproducts of pyrolysis (Table 5c).

### Liquefaction

Liquefaction, a thermochemical pretreatment process that converts organic material to bio-oil, is supposed “to mimic the natural geological processes thought to be involved in the formation of fossil fuel” in just a matter of hours or even minutes.<sup>139</sup> Conversion is conducted at 300°C, accommodating high moisture content biomass (see Pathway Map B-IV). With the help of a catalyst, the process utilizes the high activity of

**Table 5c: Endothermochemical Conversion Processes, Bioproducts, and Applications**

Pathway Process	Feedstock	Chemical Product*	Application
Pyrolysis	dry biomass	bio-oil	intermediary feedstock for hydroprocessing
		charcoal phenol-formaldehyde resins, residue	animal feed or fertilizer
Liquefaction	wet biomass	bio-oil residue	intermediary feedstock for hydroprocessing animal feed or fertilizer
		carbon dioxide (CO <sub>2</sub> )	reuse in algae cultivation process
Gasification	residue	ethanol, methanol hydrogen	biofuel hydrogen ICE (internal combustion engine) or fuel cell
		dimethyl ether (DME)	low-Btu or medium-Btu gas
		combustible gas	FT diesel; solvent, aerosol, oxygenate, intermediate for monomers and polymers

\* For detailed list of bio-based products, see table in Wang et al. (2007:1203) or Kamm and Kamm (2007:185).



water in subcritical conditions to decompose biomass materials down to those with a higher energy density or higher value chemicals.<sup>140</sup>

Liquefaction can be employed to convert the wet biomass (≥60 percent moisture) without first reducing the moisture content, thereby avoiding energy-intensive drying of biomass.<sup>141</sup>

The oil product of liquefaction can be treated to yield green diesel or green jet fuel. Process residue can either be burned (i.e., exothermal direct combustion) or converted (e.g., via fermentation) into animal feed or fertilizer. Byproducts include CO<sub>2</sub> and some recalcitrant residue.

Bio-oils produced by the thermochemical pretreatment processes of pyrolysis and liquefaction need to be upgraded before they can be used as renewable hydrocarbon biofuels. Hydroprocessing is emerging as a promising path for treating bio-oil.

**Environmental Impacts of Thermochemical Conversion**

The primary environmental issues related to thermochemical conversion are catalyst usage and energy demand.

**Catalysts**

The potential environmental impact from commercial-scale usage of conversion catalysts (whether metal, carbonaceous, alkali, etc.) needs to be considered, especially in cases where conversion efficiency is improved with higher quantities of the catalyst.

**Energy**

Liquefaction, and possibly supercritical water gasification, have an advantage over other conversion processes in that by accommodating

biomass with high moisture content, the algae-to-biofuel production pathway can bypass energy-intensive drying processes, potentially improving the energy balance of the production pathway. Depending on the circumstances, certain combinations of processes—such as the pairing of liquefaction with hydroprocessing to convert high-moisture biomass—could require less energy than a single-step conversion process that requires low-moisture biomass.<sup>142</sup>

In addition, while the use of syngas as a feedstock for the FT synthesis is a well-established gasification practice producing FT diesel, FT synthesis has been widely criticized as a particularly unsustainable fuel production practice because of its negative energy balance.

**HYDROPROCESSING**

Hydroprocessing uses a combination of heat and pressure in the presence of catalysts to upgrade a crude, intermediary feedstock to a market-ready fuel product.<sup>143</sup> Both algal oil and bio-oil can be accommodated by hydroprocessing, which upgrades the oil feedstock to a high-quality, market-ready hydrocarbon biofuel such as green or renewable diesel, jet fuel, gasoline, or other light fuel (Table 5d; see Pathway Map B-IV).<sup>144</sup>

**Conversion Processes and Characteristics**

Because the oil feedstock needs upgrading before it is fit for distribution, hydroprocessing strips it of impurities. The key components of upgrading are catalytic purification (by hydrodeoxygenation, hydrodenitrogenation, hydrodesulfurization, and hydrodemetallization) and hydrogenation through catalytic hydrocracking. Catalytic purification and hydrocracking are together known as hydroprocessing. Water, carbon dioxide, and hydrogen are the main process byproducts.

**Table 5d: Hydroprocessing Bioproducts and Applications**

Pathway Process	Feedstock	Chemical Product	Application
Hydroprocessing	bio-oil or algal oil	renewable fuel	green diesel, green jet fuel, green gasoline
		carbon dioxide (CO <sub>2</sub> ), water	reuse in algae cultivation process

### ***Environmental Impacts of Hydroprocessing***

The primary environmental issues related to thermochemical conversion are water usage and energy demand, as well as byproduct management.

#### **Water and Energy**

A big challenge in hydroprocessing is dealing with the impurities found in the oil feedstock.<sup>145</sup> Hydroprocessing potentially requires large quantities of water and energy to implement the purification and hydrocracking processes. In some cases, more water and energy may achieve better rates of production. Water and energy demand will thus depend largely on the level of impurities found in the oil feedstock.

#### **Byproducts**

As with any production process that utilizes fossil fuel-based energy for facility operations, increased usage of water and energy will also lead to increased CO<sub>2</sub> emissions. This becomes an environmental concern unless the hydroprocessing facility is co-located with the algae cultivation facility (or other outlet) for continuous recycling of CO<sub>2</sub>.

Byproduct hydrogen can be continuously recycled through the deoxygenation and catalytic hydrocracking processes. The recycling, release, or treatment practices used in managing process water, CO<sub>2</sub>, and extracted impurities (nitrogen, sulphur, metals) will play a role in determining the sustainability of hydroprocessing.

### **NONFUEL PRODUCTS**

A general overview of nonfuel products is presented here in terms of high-value coproducts, low-value coproducts, and byproducts. Environmental issues are discussed with respect to low-value coproducts and byproducts.

#### ***High-value Coproducts***

Food supplements and vitamins are the most well-known algae products. Other high-value coproducts include animal feed, pigments, and fine chemicals.

#### **Animal Feed**

The algal biomass remaining after extraction still contains significant amounts of protein and starch

as well as vitamins and minerals. Algae have been used as a component of aquaculture feed stocks for decades, including for fish, abalone, and bivalve mollusk. More recently, the biomass has been tested for use as an animal feedstock, particularly for cattle and sheep.

#### **Nutraceuticals**

Nutraceuticals are part of a health food industry, unregulated by the FDA, which consist primarily of dietary supplements derived from bioactive chemicals. These supplements include antioxidants, fatty acids, and herbal-based compounds that are extracted from fruits, nuts, fish, algae, and plants.

The main chemicals extracted from algae currently being commercialized or under consideration for commercial extraction are carotenoids, phycobilins, fatty acids, polysaccharides, vitamins, sterols, and biologically active molecules for use in human and animal health.<sup>146</sup> Many of these products have an extremely high market value. For instance, carotenoids, particularly beta-carotene and astaxanthin, command \$600/kg and \$3,000/kg, respectively. Long chain polyunsaturated fatty acids—arachidonic acid, eicosapentaenoic acid (EPA), and docosahexaenoic acid (DHA), the latter two of which are omega-3 fatty acids—are of great value to the pharmaceutical industry as supplements to prevent cardiovascular disease. Estimates of the potential U.S. market for these acids range up to \$70 million.

More specifically, algae have been used in anticoagulants, antibiotics, antihypertensive agents, blood cholesterol reducers, and antitumorigenic agents. There is potential for many additional algal products that could be marketed to the nutraceuticals industry.

#### **Pigments and Cosmetics**

Each algal species possesses pigments for photosynthesis. These pigments, the varying presence of which determines the color of an algal cell, are also used in the food and cosmetics industry.<sup>147</sup> Algae are used in thickening agents, water-binding agents, and antioxidants, as well as in iridescent pigment and packaging for cosmetics. Carrageenan (extracted from red seaweeds), for

example, contains proteins and vitamins beneficial for skin as either emollients or antioxidants.

### **Fine Chemicals**

Fine chemicals are pure, single-chemical substances that are commercially produced with chemical reactions into highly specialized applications. These chemicals tend to be custom produced in smaller quantities than in standardized, large industrial processes. Fine chemicals produced can be categorized into active pharmaceutical ingredients and their intermediates, biocides, and specialty chemicals for technical applications. Microalgae produce a large range of natural fine chemicals, such as pigments, polysaccharides, multiple unsaturated fatty acids, and other bioactive components. These products are applicable as natural ingredients in food products and supplements, pharmaceutical products, and cosmetics.

Some experts recommend that greater attention should be given to linking algal oil production to high-value coproducts and services.<sup>148</sup> The NREL ASP report concluded that only large byproduct markets, such as for animal feed, could be realistically considered for coproduction with biofuels and that the advancement of commercial-scale algae cultivation would likely be realized through specialty foods and animal feeds coproduction, rather than through the development of algal biofuel production.<sup>149</sup>

### **Low-value Coproducts**

Coproducts vary from one conversion process to another. Coproduct quantity, toxicity, and usability will be determining factors in the environmental impact they may have. Common low-value coproducts include, acetic acid, acetone, glycerol, and methane. With the exception of glycerol, which accumulates in algae and is a coproduct of transesterification, these coproducts are outputs of fermentation.

#### **Acetic Acid**

Acetic acid ( $\text{CH}_3\text{COOH}$ ) is a colorless, corrosive liquid, the vapor of which causes irritation to the eyes, nose, and throat and congestion to the lungs. Although acetic acid is an important industrial chemical used in the production of polyethylene terephthalate (PET) for use in

making soft drink bottles, at commercial scale it is an undesirable byproduct because of its limited niche applications, primarily in the production of vinyl acetate monomer, acetic anhydride, and ester production.<sup>150</sup>

#### **Acetone**

Acetone ( $\text{CH}_3\text{COCH}_3$ ) is a colorless, volatile, flammable liquid that is found naturally in the environment and in small quantities in the human body. According to the EPA, although human and animal carcinogenicity data are insufficient, to date there are no epidemiological studies demonstrating an association between exposure to acetone and increased risk of cancer.<sup>151</sup>

Acetone evaporates rapidly, is completely soluble in water, and is also consumed by microorganisms; thus it does not build up in soil, animals, or waterways, but in larger quantities may pose a significant risk of oxygen depletion in aquatic systems.<sup>152,153</sup> As an industrial chemical, it is used primarily in the production of plastics, drugs and fibers; as paint thinner or cleaning fluid; or as a solvent, drying agent, or feedstock. There is also ongoing debate as to whether acetone is a plausible automotive fuel additive for improving fuel economy and engine life.

#### **Glycerol**

Glycerol (also, glycerin or glycerine) is a colorless, liquid, sugar alcohol. The glycerol substructure is a central component of many lipids. It is a primary coproduct of the transesterification process to create biodiesel. Glycerol is considered toxic to humans, both as a liquid and in particulate form through inhalation. It also has toxic effects in the aquatic environment, negatively affecting a broad range of species from zooplankton to fish.

Glycerol can be refined for use in detergents, drugs, antifreeze, and some fine chemical industry uses. Unused glycerol is currently disposed of by incineration, which is a low-value use because of poor energy conversion and pollution generation. Because of the growing demand (and legislative mandate for) biofuels, there is a great deal of ongoing research to find high-value uses for glycerol. Possible future uses are as a feedstock for propylene glycol and replacing a petroleum

feedstock, as monomers for plastic production, and for powering microfuel cells for cell phones and other small devices. Another possible application of glycerol could be as a feedstock for algal biomass production; however, this is very dependent on the algal species and the parameters under which the algae are cultivated.<sup>154</sup>

### **Methane**

Methane (CH<sub>4</sub>), a colorless, nontoxic, highly flammable hydrocarbon gas, is the principal component of natural gas and a relatively potent GHG that remains in the atmosphere for approximately 9–15 years.

Methane can be burned as a fuel in a gas turbine or steam boiler to generate electricity. The burning process (burning CH<sub>4</sub> + O<sub>2</sub> = CO<sub>2</sub> + H<sub>2</sub>O) produces less carbon dioxide for each unit of heat released, as compared to other hydrocarbon fuels. Replacement of natural gas methane with biogas methane (from algal biomass) may have significant GHG savings.

The relative abundance of methane and its clean burning process makes it a very attractive fuel; most algal-fuel conversion until quite recently was in the form of methane. In its natural gas form, methane is transported from its source by gas pipeline (or by carrier ships, known as LNG carriers, designed for transporting liquefied natural gas).

### ***Byproducts***

Byproducts will vary from one production process to another. As an example, gasification tar is discussed here in brief. There are also other liquid (acids), gaseous (CO<sub>2</sub>), and solid byproducts of relevance to environmental sustainability of biofuel production.

### **Gasification Tar**

The thermochemical gasification process is inevitably accompanied by the formation of tar, a complex mixture of hydrocarbons.<sup>155</sup> Although residual tar (and heavy metals) is known to impact the long-term operational reliability of a gasification unit and methods for destroying the majority of tars have thus been developed, the environmental impacts of commercial-scale

accumulation of gasification tar are not widely addressed in current literature.

Efforts to minimize, destroy, or convert gasification tar in commercial-scale gasification units could contribute substantially to the environmental viability of gasification.<sup>156</sup> The environmental concerns and unknowns of tar and other byproducts should be calculated and considered in relevant impact assessments, including energy inputs for their management and disposal.

## Summary of Environmental Issues Related to Oil and Residue Conversion

There are a few environmental benefits, concerns, and unknowns that can be identified with respect to algal oil and residue conversion. The criteria for sustainable conversion should consider potential energy demand and variety and usability of nonfuel products.

The environmental impacts of oil and residue conversion will depend on the conversion processes implemented, maximum biomass moisture content permitted, energy inputs and outputs, types and quantities of catalyst(s), as well as expected nonfuel coproducts and byproducts and their toxicity or reusability. Such variability in potential impacts exhibits the need for greater analyses of entire production pathways to determine the true energy balance and environmental costs and overall prospects for sustainability.

### ENVIRONMENTAL BENEFITS

- ▶ Reuse of gases and waste heat in conversion processes
- ▶ Use of low-value coproducts glycerol, certain acids, and recalcitrant biomass residue

### ENVIRONMENTAL CONCERNS

- ▶ Processes may be very energy intensive
- ▶ Improved process efficiencies are often achieved via higher resource consumption (water, energy, catalysts)
- ▶ Wastewater (when an output) could require treatment
- ▶ Low-value bioproducts not reused by the production system would need to be managed, stored, and/or disposed

### ENVIRONMENTAL UNKNOWNNS

- ▶ Energy balance
- ▶ Impact of tar and other conversion byproducts at scale



## CHAPTER 4

# Conclusions and Recommendations for Algae Biofuel Production

**A**lgal biomass holds the potential to meet a significant portion of our global fuel demand, and with 140 billion gallons of motor fuel consumed every year in the United States alone—and more than 320 billion gallons consumed worldwide—the potential demand for cleaner, renewable algae biofuels is enormous.<sup>157, 158</sup> By assessing production pathways, processes, inputs, and outputs, many relationships have been identified between one process and another and between those processes and the environment. These relationships have been summarized in terms of the environmental benefits, concerns, and unknown impacts of commercial-scale algae-to-biofuel production on water, land, soil and biodiversity, air, and energy.

### ENVIRONMENTAL BENEFITS

Of the known positive externalities resulting from algae-to-biofuel production, there exist the following environmental benefits:

- ▶ Wastewater can be biologically treated.
- ▶ Diverse water types and qualities can be accommodated
- ▶ Pathways are interchangeable and could potentially be designed according to local or regional environmental conditions, climate conditions, and availability of resources
- ▶ Process outputs are often recyclable as inputs to adjacent processes
- ▶ Environmentally responsible biofuel production minimizes water, energy, and land usage and reduces the degradation of water, soil, biodiversity, and air quality, with the potential to improve their condition
- ▶ Linking algae biofuel production with other existing industries, where the waste of one industry becomes the feedstock for another (i.e., industrial ecology), could improve resource management and minimize the ecological footprint of a production pathway

### ENVIRONMENTAL CONCERNS

Of the known negative externalities resulting from algae-to-biofuel production, there exist the following environmental concerns:

- ▶ Environmentally responsible alternatives could be passed over in favor of more cost- or time-efficient processes
- ▶ Water usage in one process could affect energy usage in subsequent processes
- ▶ Certain inputs may increase the scope of a pathway to include additional processes (i.e., chemical recovery or wastewater treatment), which could increase energy demand
- ▶ Extensive land transformation and changes in water and air quality could impact local or regional hydrology, native habitats, and migratory patterns
- ▶ Materials toxicity could have long-term impacts on biodiversity, soil and water quality, and/or aquifer recharge
- ▶ Processing facilities that are not co-located will require increased storage and transport, and thus increased land and energy usage

Of the five core environmental issues, energy and water usage surface as having the greatest potential variable influence, whether positive or negative, at commercial-scale production. Energy and water represent significant inputs or outputs in most production processes, and their respective quantities and impacts could vary dramatically from one pathway to another. Although the degree of impact from one pathway to another cannot be determined without a more thorough analysis, it can be foreseen that any two given pathways could have very different effects, both anticipated and unknown.

### ENVIRONMENTAL UNKNOWNNS

With respect to externalities resulting from algae-to-biofuel production, there exist the following environmental unknowns:

- ▶ Potential for CO<sub>2</sub> abatement
- ▶ Impacts of indirect water inputs and land usage
- ▶ Long-term effects of direct water, land, and energy usage on ecosystem health and watershed sustainability
- ▶ Overall energy balance of production
- ▶ Impacts of increased quantities of process byproducts

- ▶ Potential impact of environmental contamination by genetically modified algae or “renewable oil spills”

Identified environmental unknowns tend to highlight the need for both production data about system demands on water, land use, and energy, and a greater awareness of the long-term impacts of direct and indirect process inputs. Many of these unknowns are due simply to a lack of observable data, limited by the number of pilot- and commercial-scale projects.

Having assessed algae cultivation and fuel production methods and identified these critical environmental challenges, it can be concluded that the environmental benefits provided by algae-to-biofuel production have the potential to make significant contributions to a sustainable biofuels industry. However, the way water, nutrients, land, light, and other inputs are supplied and managed could have a significant effect on both the energy balance of a production pathway and the persistence of environmental quality. Therefore, in order to establish sustainable production pathways, associated environmental concerns and unknown impacts must be addressed through relevant technologies and policies that are developed as the algae biofuel industry moves forward. In light of this definitive need to improve the industry’s environmental sustainability, next steps for policy and industry have been outlined in the following section on recommendations.

### Recommendations

Concerns regarding long-term replacements for petroleum, environmental issues, and national security have helped to ignite investments in all types of clean energy, including algae fuels research. Although most work is focused on research and development and small demonstration systems, several companies are pursuing production-scale operations and testing their products for commercial use. Several environmental challenges will persist in the production of algae biofuels, however, until sustainable production processes are fully established.

In the near term, there will be trade-offs between production pathways that are economically feasible and those that are environmentally sustainable, especially with respect to algae cultivation and biomass harvesting. Until major biological and technical

barriers are overcome, the industry may need to engage otherwise underutilized land, water, and nutrient resources, biological services, and nonfuel coproducts to make algae biofuels economically viable. In order to develop and commercialize a sustainable product in the long term, research and development of algae biofuel production will need to closely address the externalities of scaled biofuel production processes—with respect to both direct and indirect land, water and energy inputs, chemical usage, land transformation, and materials fabrication and toxicity—and their potential impacts on the environment.

### **POLICY MAKERS**

From a regulatory and policy standpoint, there are several tasks that would push commercialization of the algal biofuel industry in a sustainable direction. The following tasks should be undertaken at a minimum:

- ▶ Clarify roles and responsibilities within government agencies
- ▶ Encourage subindustry collaboration
- ▶ Begin life cycle analysis (LCA) at the fuel product design phase
- ▶ Reenforce need for conducting environmental impact statements (EIS)
- ▶ Develop a regulatory roadmap
- ▶ Inventory all regulations and guidelines (information resource)
- ▶ Specify sustainability metrics and industry standards<sup>159</sup>
- ▶ Adopt international standards for sustainable biofuels<sup>160</sup>

### **INDUSTRY**

To inform investment and policy decisions, greater research is needed on issues that will have environmental implications for algae biofuel processes. Industry should proactively address the following issues for their respective technologies, keeping in mind the relationships they hold with other processes of algae-to-biofuel production.

- ▶ Conduct technoeconomic analyses
- ▶ Conduct a water balance
- ▶ Conduct energy and carbon balances
- ▶ Consider environmental impacts to native habitats in proximity to production and processing facilities; adopt low-impact development, operations and maintenance practices

- ▶ Chemical recovery and nonchemical substitutes for biomass recovery
- ▶ Materials toxicity and resource consumption for materials fabrication
- ▶ Begin life cycle analysis (LCA) at the fuel product design phase
- ▶ Encourage transparency of process inputs and outputs
- ▶ Improve understanding of how relationships among production processes define resource consumption and management (e.g., relationship between water inputs and heat or electric energy inputs)

Establishing sustainable, scaled production pathways will require policy makers and the algae biofuel industry to leverage the environmental benefits and address the concerns and unknown impacts caused by unsustainable practices. Measurement of process inputs and outputs will be one of the keys to determining pathway sustainability, and collaboration among subindustries will help focus and unite such efforts.

The environmental benefits provided by algae-to-biofuel production have the potential to make significant contributions to a sustainable biofuels industry. However, associated environmental concerns and unknown impacts must be addressed through relevant technologies and policies to ensure the algae biofuel industry scales up in a consistent and beneficial manner.

## Appendices

### Appendix A: Historical Overview of Algae Biofuel Research and Development

The fossil fuels we use today to power our world are the fossilized ancestors of life on this planet. Billions of years ago, algae excreted oxygen that formed the ozone layer and the oxygen we breathe. The lipids and hydrocarbons produced by these organisms created the black oil we use today.

The concept that algae can be used to create useful products is nothing new. For more than 500 years, man has collected micro- and macroalgae for food. Over the past 50 years, algae research has transitioned its focus from growing food to producing energy, cleaning waterways, and even supporting life in space. Recent algae-based product and service applications have expanded to include agricultural feedstock, aquaculture, fertilizer, cosmetics, pigment, toxic waste remediation, and designer fuels.

#### Pharmaceuticals, Nutraceuticals, and Health Food

The first pharmaceutical derived from microalgae, known as chlorellin, was extracted from *Chlorella* in Japan during World War II.<sup>161</sup> This discovery had a profound impact on the future of the pharmaceutical industry. Today, *Spirulina* and *Dunaliella* are the primary algal species used in food and nutritional products. *Spirulina* was discovered to be an excellent source of protein consumed by native African tribes. Its commercialization in 1975 by Larry Switzer led to the creation of Earthrise Farms in southern California, the largest microalgae facility in the world.<sup>162</sup> *Dunaliella* did not have the same success in the United States. The microalgae facilities used to cultivate *Dunaliella* to extract glycerol and beta-carotenes proved to be less expensive to operate in Australia and are no longer operational in the United States.<sup>163</sup>

#### Post-World War II Biofuels, 1950s

The discovery that microalgae can be transformed into biofuel is a concept that evolved from the large-scale, algae-based food production research of the early 1950s. In 1951, Arthur D. Little developed a pilot plant to study algal food production for the Carnegie Institute in Washington DC.<sup>164</sup> Whereas his study of algae for large-scale food production was considered unsuccessful, Little's documentation of the fundamental characteristics of algae became the basis for all future research.

#### Algae-based Wastewater Treatment, 1960s

R.L. Meier conducted the first study of using algae for energy production in 1955. Subsequently, during the early 1960s, William Oswald and Clarence Golueke introduced the concept of growing microalgal biomass in large raceway ponds to produce methane gas.<sup>165</sup> The concept of using lipids derived from algae to create methane gas is the basis of modern day microalgal fuel research.

#### Space Travel and Algae as a Source of Life Sustaining Nourishment, 1959–1962

Between 1959 and 1962, Professor Jack Myer at the University of Texas at Austin and Professor Herb Ward at Rice University began conducting algae-related bioscience experiments for the U.S. Air Force's School of Aviation Medicine (SAM) in San Antonio, Texas. Because algae release oxygen from water and utilize light efficiently, there was an interest in studying algae for use in life-support systems for space travel.<sup>166</sup> SAM conducted tests on the efficiency of microalgae to produce oxygen in closed-loop systems.

The next major breakthrough to come out of the SAM program was the "Algatron," a novel algal photobioreactor designed to mechanically rotate the culture to cultivate algae with a high rate of efficiency. The bowl-shaped device consisted of a centrifuge that would spin a thin layer of algae at 300 rpm onto the walls of the Plexiglas reactor. The Algatron is the highest-efficiency culture density photobioreactor ever produced.<sup>167</sup> Three Algatrons, each one-meter in diameter, are suggested to be able to support one person with 1,600 calories of food

per day.<sup>168</sup> The invention, though never realized outside the laboratory, solidified algae as a source of life sustaining nourishment.

Microalgae research was later abandoned after SAM transformed into the National Aeronautics and Space Administration (NASA), which directed its attention to physical and chemical methods of recycling water and oxygen.<sup>169</sup>

### **NREL Aquatic Species Program, 1978–1996**

The energy crisis of the 1970s created a renewed interest in biofuel research, which led to the creation of the U.S. Department of Energy's Aquatic Species Program (ASP). From 1978 to 1996, ASP studied the potential of using algae to create first hydrogen and then liquid transportation fuel. The primary focus of the program was to produce a biodiesel from high-lipid-content algae grown in open ponds using waste carbon dioxide from coal-fired power plants.<sup>170</sup> Over nearly 20 years the ASP explored all aspects of microalgae research, resulting in a solid foundation for developing fuel from algae technology. The ASP received a total of 25 million dollars during its 18-year history.<sup>171</sup> In 1996, the remaining cultures were transferred to the Center for Marine Microbial Ecology and Diversity at the University of Hawaii. The total amount of algae research done following the termination of the ASP is relatively small because of the lack of funding and growing interests in cellulosic ethanol.

By the early 1990s, “the desirable traits for biodiesel (high productivity and high lipid content) were found to be mutually exclusive” in the algae cultivation process. The program subsequently concluded that mutagenesis or genetic engineering would be necessary to manipulate the algae to produce algal strains with “constitutively high” TAG lipid levels and thus a more efficient feedstock for commercial-scale biofuel production.<sup>172</sup>

### **Japan's NEDO-RITE Optical Fiber Bioreactors, 1990–2000**

As major advances in the biochemistry of photosynthetic microbes were made in the 1980s and 1990s, research has broadened in scope—covering topics from species-specific photosynthetic capacities, to novel photobioreactor designs, to integrated algae-based wastewater treatment and CO<sub>2</sub> abatement—yet limited in quantity because of a lack of funding. However, Japan's NEDO-RITE Optical Fiber Bioreactor project brought algae back to the forefront of scientific research. The 10-year program beginning in 1990 obtained the support of more than 20 private companies, laboratories, and academic institutions, and more than 200 million U.S. dollars in funding.<sup>173</sup> The Japanese closed photobioreactors were designed to use optical fibers, which would diffuse light into the reactors creating a controlled environment for the algae to grow.<sup>174</sup> This control would allow for fixed environmental conditions, such as increased levels of CO<sub>2</sub>, which could then maximize the quality and yield of algae obtained. Although the Japanese set out to develop a technology that could replace coal with algal biomass, the program was largely unsuccessful and the U.S. National Renewable Energy Laboratory has referred to the program as a “total failure,” explaining that algae costs exceeded ~\$1000/m<sup>2</sup>.<sup>175</sup> The Japanese RITE program competition in 2000 marked the last major international algae experimentation to date; however, numerous companies, start-up companies, and researchers continue to push for the commercialization of algal biofuels today.

### **Recent Progress, 1996–2009**

After the Department of Energy's ASP was discontinued, activity slowed in the United States in developing algae-based biofuels. However, algae have been crucial in helping scientists gain a greater understanding of photosynthesis and the effects of anthropogenic disturbance to the natural environment, which have led to many advances resulting from the combination of biological and engineering knowledge.<sup>176</sup> These advancements have included direct and indirect food supplements, the treatment of polluted effluents, and the refinement of more efficient algae-based fuels.

Most recently, universities, as well as start-up companies funded by venture capitalists, have been advancing current knowledge through pilot studies in the cultivation and downstream processing of algae.

As oil prices skyrocketed in 2008 and several airline carriers filed for bankruptcy, the race to develop alternative fuel for passenger planes heightened. To help airlines cut fuel costs and reduce carbon emissions, fuel companies



and start-ups have begun partnering with airlines to test a new generation of biofuels that have ventured to include algae.

On January 7, 2009, Continental Airlines Inc. successfully flew a Boeing 737 twin-engine jet, powered partly by a mix of 50 percent kerosene and a blend of algae- and jatropha-based fuels.<sup>177</sup> The two-hour test flight over Houston was the first by a U.S. carrier; no passengers were on board.

## Appendix B: Discussion of Genetically Modified Algae

Industry sources have varying concerns over environmental threats of genetically modified organisms (GMOs). Because of their extreme dependence on specific environmental conditions, transgenic algae may be incapable of surviving in the wild.<sup>178,179</sup> This may be difficult to prove, however, because of the lack of GMO monitoring and data to prove either case as fact. According to a 2008 policy paper published in *Science*,<sup>180</sup> better monitoring is needed of genetically engineered agricultural crops in use; and in a public statement announcing the study of federal regulation of GMOs,<sup>181</sup> Dr. David Schubert of the Salk Institute for Biological Studies summarized the conclusion of their analyses by stating that “U.S. regulators rely almost exclusively on data provided by the biotech crop developer,” which have neither been subjected to peer review nor published in journals.<sup>182</sup>

### Industry Benefits

Transgenic yeasts and bacteria have been used at the industrial scale for over 30 years and there has been no major report to date of significant adverse consequences to the environment. This is important to note because yeast and bacteria are more prolific than algae and would likely be more difficult to subdue in an uncontrolled environment.<sup>183</sup>

Gressel (2008) states that transgenics have the potential to overcome challenges to efficient cultivation related to algae survival, growth rate and lipid content, CO<sub>2</sub> enrichment (fixation rate), light penetration, and temperature, as well as tolerance of high-stress harvesting methods.<sup>184</sup> Transgenic algae modified to reduce the size of a cell’s solar antenna (known as *tla1*, or truncated light harvesting chlorophyll antenna) could increase the cell’s photosynthetic capacity even during the height of daytime exposure.<sup>185</sup> A development such as this could have significant impact on the efficiency of algal biomass production.

### Environmental Concerns

Primary concerns with GMO use are related to unknown threats to the proliferation of biodiversity, alteration or displacement of native species, contamination of organic agricultural crops, and long-term effect on human health.

A report released in 2004 by the Union of Concerned Scientists expresses concern over the potential implications of traditional plants becoming contaminated with genetically engineered elements. “Once genetically engineered plants are released into the environment,” states the report, “historically preserved and heirloom seed strains are forever affected.” As a result of this contamination, diverse agricultural economies could be at risk of losses. An example given by the report suggests the potential for farmers to lose their organic certification if their organic crops become contaminated with genetically engineered pollen.<sup>186</sup> Although there is no evidence that this scenario applies to algae, there is also no evidence that it would not happen. Therefore, if transgenic algae are to be mass-cultivated, appropriate mitigation strategies should be generated and protection should be considered for all stakeholders.<sup>187</sup>

Although it may be difficult to quell curiosity about GMO applications, some experts have expressed concern that the future of the algae biofuel industry as a whole rests too heavily on the public perception of GMOs. The first step may be to exhaust research options for naturally existing varieties and having a complete understanding of what attributes are needed before delving into genetic modification.<sup>188</sup> Some argue, however, that genetic modification is the future of algal biofuels; and since the goal is not to create a food or pharmaceutical product, there should be less

emphasis on the potential impacts of GMO algae. After all, commercial-scale industries have been using GMOs for decades.

### **GMO Regulations and Policy in Relation to Algae Cultivation**

Certain U.S. states or counties may limit or restrict the cultivation of transgenic algae under GMO permitting and local- or state-based GMO regulations, further restricting land options for algae cultivation.<sup>189</sup> This points to the value in addressing GMO concerns early on, before they become a larger barrier for industry progress.

#### ***GMO Permitting***

Since 1986, three federal agencies, the Environmental Protection Agency (EPA),<sup>190</sup> the Food and Drug Administration (FDA),<sup>191</sup> and U.S. Department of Agriculture's Animal and Plant Health Inspection Service (USDA-APHIS)<sup>192</sup> have been responsible for regulating genetically engineered crops. No federal laws have been passed to specifically regulate genetically engineered crops; however, there has been much criticism of the USDA-APHIS permitting processes for GMOs.<sup>193</sup> Concerns have been expressed for a number of issues, including the classification of GMOs under the generic category "novel protein"<sup>194</sup> and the use of speculative language in environmental assessments.<sup>195</sup>

#### ***Local and State-based GMO Regulation***

Efforts to cultivate GMO algae may encounter barriers to achieving public acceptance at the local and state level. Several state legislatures have attempted to subdue this growing resistance to GMOs by passing "preemption laws," restricting the ability of political subdivisions to regulate seeds and plants with existing or future ordinances, rules, or regulations.<sup>196</sup> Very few state governments have attempted to restrict genetically engineered plants or plant parts from being grown or transported within state lines. Mounting support for farmers in the form of legal protection from liability, however, highlights the difficulty in confronting biotech conglomerates and large-scale agriculture.

GMO regulations may not apply to algae if their commercial production is termed an industrial activity rather than an agricultural one. The type of cultivation system employed may also determine the applicability of agricultural GMO policies

## Acronyms

AIWPS	Advanced Integrated Wastewater Pond System®
ASP	Aquatic Species Program
ATS	Algal Turf Scrubber®
DOE	U.S. Department of Energy
FAO	U.S. Food and Agriculture Organization
HRT	Hydraulic Retention Time
NRDC	Natural Resources Defense Council
NREL	National Renewable Energy Laboratory
PBR	Photobioreactor

## Glossary of Terms

### Algae

A large and diverse group of simple organisms, ranging from unicellular to multicellular forms. Algae are photosynthetic, like plants, yet “simple” because they lack the many distinct organs found in plants. Many are photoautotrophic, although some are mixotrophic, deriving energy both from photosynthesis and organic carbon uptake. Some unicellular species rely entirely on external energy sources and have limited or no photosynthetic apparatus. However, nearly all algae have photosynthetic machinery that allows them to produce oxygen as a byproduct of photosynthesis.

### Biodiversity

The variation of life forms (flora, fauna, bacteria, etc.) within a given ecosystem. Biodiversity is often used as a measure of ecosystem health.

### Biofuel

A solid, liquid, or gaseous fuel obtained from relatively recently lifeless biological material that is different from fossil fuels, which are derived from long-dead biological material. Many biofuels are biodegradable.

**First-generation biofuels** are made from sugar, starch, vegetable oil, or animal fats using conventional technology. The basic feedstocks for the production of first-generation biofuels are often grains, which yield starch that is fermented into bioethanol, or seeds, which are pressed to yield vegetable oil that can be used in biodiesel.

**Second-generation biofuels** use biomass to liquid technology, including cellulosic biofuels from nonfood crops, including waste biomass, the stalks of wheat, corn, or wood. These biofuels are inherently more efficient than first-generation technologies because they use more of the plant to produce fuel. They are also known as “advanced biofuels.”

**Third-generation biofuels** are derived from algae. They are also known as “advanced biofuels.”

### Biological wastewater treatment

The purpose of biological treatment is purification of contaminants. Algae and bacteria cells feed on the organic materials in the wastewater, thereby reducing its nutrient, toxin, and organic material (BOD) content. Through their metabolism, the organic material is transformed into cellular mass (e.g., algal biomass). Biological wastewater treatment is achieved by mechanical means or by biological filters, such as vegetation or constructed wetlands, to accomplish what is generally called secondary treatment.

### Biomass

As a renewable energy source, biomass refers to living or recently dead biological material that can be transformed into fuel or used for industrial production. Industrial biomass can be grown from numerous types of plants and algae.

**Bioproduct**

A product derived from the processing of biological matter, such as biomass.

**Byproduct**

A secondary or incidental product derived from a manufacturing process, chemical reaction, or biochemical pathway, and is not the primary product or service being produced. A byproduct can be useful and marketable, or it can have a negative ecological impact. For the purpose of this report, the term refers to a process output with little or no value (in the existing market) that must be disposed of (e.g., CO<sub>2</sub>, tar, certain acids), or to which pretreatment is required in order to reclaim value or viable reuse (e.g., wastewater).

**Carbon fixation**

Carbon fixation is a process found in organisms that produce their own food (autotrophs), usually driven by photosynthesis, whereby carbon dioxide is changed into organic materials (sugars).

**Coproduct**

A product or service produced in conjunction with another, often to balance the economics of production. For the purpose of this report, coproducts refer to process outputs (e.g., bio-oil, biofuel, animal feed) that have an existing or emerging application (market).

**Energy balance**

An aggregate presentation of all activities related to energy, except for natural and biological processes. An energy balance can be quantified by comparing the energy inputs and losses, as in energy consumed or released (measured as total energy or, more commonly, total fossil fuel energy), to yield a specific product (e.g., energy balance of an algae-based biofuel would include all energy inputs and outputs, from algae cultivation to its market-ready product).

**Feedstock**

A raw material that came from nature and is in an unprocessed or minimally processed state.

**Flocculant**

Typically a high-molecular-weight polymer of natural or synthetic origin, used as an additive in the harvesting process to coerce algal cells to coagulate into larger clumps, enabling easier extraction of biomass from the culture medium.

**Genetically modified organism (GMO)** See “transgenic.”

**Greenhouse gas (GHG)**

Gases in the atmosphere that absorb and emit radiation within the thermal infrared range. High concentrations of these gases are said to cause the change in the steady state temperature of a planet. Common greenhouse gases in the earth’s atmosphere include water vapor, carbon dioxide (CO<sub>2</sub>), methane (CH<sub>4</sub>), nitrous oxide (NO), ozone, and chlorofluorocarbons (CFC).

**Hydraulic retention time (HRT)**

A measure of the average length of time that a soluble compound (e.g., algal culture) remains in a reactor (e.g., pond, photobioreactor).

**Hydrocarbon (biofuel)**

An organic compound consisting entirely of hydrogen and carbon and no oxygen. Hydrocarbons are one of the earth’s most important energy resources. The predominant use of hydrocarbons is as a combustible fuel source—the main components of gasoline, naphtha, and jet fuel. They are also the main source of the world’s electric energy and heat sources (such as home heating). All hydrocarbon combustion reactions produce carbon dioxide and water. Common hydrocarbon fuels are “green” or “renewable” gasoline, diesel, and jet fuels, as well as methane (CH<sub>4</sub>), ethane (C<sub>2</sub>H<sub>6</sub>), and propane (C<sub>3</sub>H<sub>8</sub>).

**Life Cycle Analysis (LCA)**

A life cycle analysis (also known as life cycle assessment, ecobalance, and cradle-to-grave analysis) is the investigation and valuation of the environmental impacts of a given product or service caused or necessitated by its existence.

**Lipids**

Hydrocarbons with higher energy density than that of other plant components, such as carbohydrates. Lipids are water-insoluble fat molecules. There are several types of lipids; the primary lipids involved in energy in algal cells are triacylglycerides (TAGs). See also “triacylglycerol.”

**Oxygenate (biofuel)**

Fuels that are typically made from triacylglycerides (TAGs), yielding biodiesel, or from biomass sugars and starches, yielding biobutanol and bioethanol. See also “triacylglycerol.”

**Pan-evaporation Rate**

Regionally specific data measured by the United States National Weather Service, using the four-foot diameter Class A evaporation pan.

**Photobioreactor (PBR)**

An enclosed culture vessel that is designed to utilize natural or artificial light to support photosynthesis for controlled biomass production.

**Photosynthesis**

A metabolic pathway that converts carbon dioxide into organic compounds, especially sugars, using the energy from sunlight. Photosynthesis occurs in plants, algae, and many species of bacteria. Photosynthetic organisms are called photoautotrophs, but not all organisms that use light as a source of energy carry out photosynthesis, since photoheterotrophs use organic compounds, rather than carbon dioxide, as a source of carbon. In plants, algae and cyanobacteria photosynthesis uses carbon dioxide and water, releasing oxygen as a waste product. Photosynthesis can be described by the simplified chemical reaction  $H_2O + CO_2 + \text{energy} \rightarrow CH_2O + O_2$ , where  $CH_2O$  represents carbohydrates such as sugars, cellulose, and lignin.

**Photosynthetic Efficiency**

The fraction of light energy converted into chemical energy during photosynthesis in plants and algae.

**Renewable BioFuel** Also known as green fuel. See “hydrocarbon.”

**Residue**

A bioproduct of biomass processing—the residual matter remaining after the separation of oils from biomass. Residue (also known as algae meal or mash) can be comprised of carbohydrates, proteins, nutrients, and ash, which can be processed into animal feed, fertilizer, or other nutrient-rich product.

**Sustainable Biofuel**

A biofuel that is produced in a manner that does not degrade or diminish natural or human resources and is environmentally, economically, and socially sustainable. See also “biofuel” and “life cycle analysis.”

**Transgenic (algae)**

Transgenic algae possess a gene or genes that have been transferred from a different algal species or other organism. Although DNA of another species can be integrated in an algal genome by natural processes, the term “transgenic algae” refers to algae created in a laboratory using recombinant DNA technology for the purpose of creating algae with specific characteristics.

**Triacylglycerol (TAG)**

Triacylglycerol (also known as triglycerol, triacylglyceride, or TAG) is glyceride that has been naturally esterified with three fatty acids. TAG is a type of lipid water-insoluble fat molecule that can be found in algal cells. There are several types of lipids in algae; however, TAGs are the primary lipids involved in energy storage as well as the primary type of lipid associated with biodiesel production.



## Endnotes

- 1 GreentechMedia.com, Algae Biofuel Investments Explode, September 18, 2008, accessed January 12, 2009, <http://www.greentechmedia.com/articles/read/algae-biofuel-investments-explode-1434/>.
- 2 AquaticBiofuel.com, 2008 the year of algae investments, December 5, 2008, accessed June 30, 2009, <http://aquaticbiofuel.com/2008/12/05/2008-the-year-of-algae-investments/>.
- 3 GreentechMedia.com, Solix:Another Me-Too Algae Company Raises \$10.5M, November 11, 2008, accessed at <http://www.greentechmedia.com/articles/solix-another-me-too-algae-company-raises-105m-5151.html>.
- 4 EIA 2007 statistics, located under “Finished Motor Gasoline,” accessed at [http://tonto.eia.doe.gov/dnav/pet/pet\\_cons\\_psup\\_dc\\_nus\\_mbbldpd\\_a.htm](http://tonto.eia.doe.gov/dnav/pet/pet_cons_psup_dc_nus_mbbldpd_a.htm).
- 5 Calculated from the most recent DOE EIA numbers in 2004, where the United States accounted for 44 percent of all motor fuel consumption; accessed at <http://www.eia.doe.gov/pub/international/iea2005/table35.xls>.
- 6 Chivian E. and Bernstein A., 2008, *Sustaining life: how human health depends on biodiversity*, E. Chivian and A. Bernstein (Eds.), Oxford University Press: New York, p.52.
- 7 Barsanti L. and Gualtieri P., 2006, *Algae: anatomy, biochemistry, and biotechnology*, Boca Raton: Taylor and Francis Group, LLC, p.163.
- 8 Recharge is usually expressed as an average rate of inches of water per year, similar to precipitation. In addition to precipitation, other sources of recharge to an aquifer include stream, lake or pond seepage, irrigation return flow (both from canals and fields), interaquifer flows, and urban recharge (from water mains, septic tanks, sewers, drainage ditches).
- 9 One possible means to remediate this impact is to directly inject clean water into the watershed or aquifer at a rate equal to the predevelopment rate of percolation.
- 10 Mockus S.M., Sciasly D. and Johnson R., “Algae for biofuels: current potential for commercial development,” Webinar (March 11, 2009), William Bardosh (host), the Garbrook Knowledge Resource, Advanced Biofuels Resource (ABR); Personal communication with Arthur Grossman, Carnegie Institution of Washington, December 2008.
- 11 In 2002, 51.8 percent of the total U.S. land area was used for agricultural activities; Lubowski R.N., Vesterby M., Bucholtz S., Baez A. and Roberts M.J., 2002, Major uses of land in the United States, 2002/EIB-14. Economic Research Service/USDA.
- 12 Antonios P., 2008, “Biodiversity is vital for human survival and livelihoods,” FAO Deputy Director-General says, FAO Newsroom, accessed April 6, 2009, <http://www.fao.org/newsroom/en/news/2008/1000788/index.html>.
- 13 John Spengler, director of Environmental Science and Engineering program, Harvard School of Public Health, cited in Devra Lee Davis, 2002. *When Smoke Ran Like Water*. New York: Basic Books.
- 14 NEWTON “Ask A Scientist”, Argonne National Laboratory, Division of Educational Programs, accessed 4 June, 2009, <http://www.newton.dep.anl.gov/askasci/chem00/chem00995.htm>.
- 15 van Harmelen and Oonk, 2006.
- 16 In algae cultivation, energy balance can be quantified by comparing “carbon accumulation in the new biomass with photosynthetic electron transport rates per absorbed quantum, measured both by fluorescence quenching and oxygen production. The difference between fluorescence- and oxygen-based electron flow is defined as ‘alternative electron cycling’” Wagner H., Jakob T. and Wilhelm C., 2006, “Balancing the energy flow from captured light to biomass under fluctuating light conditions,” *New Phycologist* 169(1):95–108.
- 17 The net energy gain in algae cultivation depends on “a differential temperature effect on gross photosynthesis and endogenous respiration in various plankton groups” (Andersson, Haecky and Hagstrom, 1994). For instance, “*Botryococcus braunii* is more profitable for liquid fuel production than *Dunaliella tertiolecta* based on calculating the energy balance” (Tsukahara and Sawayama, 2005); and *Phaeodactylum tricornutum* (diatom) is more energy efficient than *Chlorella vulgaris* (green algae) under fluctuating light conditions (Wagner, Jakob and Wilhelm, 2006).
- 18 Lipids are water-insoluble fat molecules. There are several types of lipids; the primary lipids involved in energy in algal cells are triacylglycerols or TAGs. Other (non-TAG) lipids present in the cell are sterols, fatty acids, oils, fat-soluble vitamins (A, D, E, and K), monoglycerides, and diglycerides. Triacylglycerol is glyceride that has been naturally esterified with three fatty acids. It is also the primary type of lipid associated with biodiesel production.
- 19 Water vapor contributes 36–72 percent of the earth’s GHGs, followed by carbon dioxide, which contributes 9–26 percent, methane 4–9 percent, and ozone 3–7 percent, Kiehl J.T. and Trenberth K.E., 1997, “Earth’s Annual Global Mean Energy Budget,” *Bulletin of the American Meteorological Society* 78(2):197–208.
- 20 van Harmelen and Oonk, 2006.
- 21 Rai et al., 2000.
- 22 Carbon fixation is a process in autotrophs (organisms that produce their own food), usually driven by photosynthesis, whereby carbon dioxide is converted into energy-rich organic carbon. Algae can also fix nitrogen, phosphorous, and some heavy metals such as copper.
- 23 Patil V., Tran K. and Ragnar Giselrød H., 2008, “Towards sustainable production of biofuels from microalgae,” *International Journal of Molecular Sciences* 9:1190; van Harmelen and Oonk, 2006.
- 24 From 1978 to 1996, the DOE Aquatic Species Program researched the production of biodiesel from algae. As part of the study, algae grown in large-scale ponds in Roswell, New Mexico, were fed exhaust gas from coal-fired power plants. By carefully controlling pH and physical conditions, researchers achieved a 90 percent utilization of the injected CO<sub>2</sub> and high algae yields.
- 25 See for example: “Algae farms can clean up power plants’ carbon”, *Billings Gazette*, September 12, 2009; [http://billingsgazette.com/news/opinion/guest/article\\_a463df08-9f54-11de-a466-001cc4c03286.html](http://billingsgazette.com/news/opinion/guest/article_a463df08-9f54-11de-a466-001cc4c03286.html)
- 26 See for example: “APS gets \$71M for carbon sequestration program”, *Phoenix Business Journal*, September 15, 2009; <http://phoenix.bizjournals.com/phoenix/stories/2009/09/14/daily36.html>.

- 27 Kadam K.L., 2001, Microalgae production from power plant flue gas - environmental implications on a life cycle basis, Prepared for the National Renewable Energy Laboratory, NREL/TP-510-29417
- 28 A study sponsored by the National Renewable Energy Laboratory (NREL) Aquatic Species Program (ASP) concluded that CO<sub>2</sub> recovery from existing emissions processes was judged to be relatively low cost in ethanol and ammonia plants, and high cost from cement plants, refineries, and power plants. (Feinberg D.A. and Karpuk M.E., 1990, *CO<sub>2</sub> sources for microalgae based liquid fuel production*, Report, Solar Energy Research Institute, Golden, Colorado, SERI/TP-232-3820); see also, Karpuk M., 1987, "CO<sub>2</sub> sources for fuels synthesis," FY 1986 *Aquatic Species Program Annual Report*, Solar Energy Research Institute, Golden, Colorado, SERI/TP-231-3071, 269-275.
- 29 Benemann, 2003.
- 30 There is expressed interest in accessing carbon dioxide by locating near fossil fuel-based power plants; according to Garbrook Knowledge Resources, there are currently 29 companies using algae to capture CO<sub>2</sub>, Mockus, Sciasy and Johnson, 2009.
- 31 Abatement contributions (percentage of available CO<sub>2</sub> utilized) by an algae cultivation system could prove insignificant to the average 500MW coal power plant that releases 3.5 million tons of CO<sub>2</sub> emissions per year. The availability of land or other environmental concerns could also dictate feasibility of co-location with industrial plants. Waltz E., (n.d.), *Genetically manipulating algae for fuel: Little green monsters or clean energy to save the world?* accessed September 23, 2008, from Plenty the World in Green, [http://www.plentymag.com/features/2008/09/gm\\_algae.php](http://www.plentymag.com/features/2008/09/gm_algae.php).
- 32 See source for specifications: Feinberg and Karpuk, 1990:32
- 33 For example, gasification is said to offer the most feasible option for capturing CO<sub>2</sub> from power generation. There are more than 140 gasification plants operating worldwide, 19 of which are located in the United States (Gasification Technologies Council, [http://www.gasification.org/what\\_is\\_gasification/environmental\\_benefits.aspx](http://www.gasification.org/what_is_gasification/environmental_benefits.aspx)). It is important to note that biomass gasification is different from the current inventory of gasifiers, which run on pet coke, coal, and refinery waste.
- 34 Sayre R., "Algal Biofuel Technology Roadmap Workshop," sponsored by U.S. Department of Energy, University of Maryland (December 9-10, 2008).
- 35 Although scientists have estimated the existence of tens of thousands of algal species, approximately 30,000 species have been identified and even fewer have been subjects of intensive culture.
- 36 In the context of biofuel production, feedstock is the raw material (input) supplied to a machine or industrial process to produce biofuel (output).
- 37 Huntley M.E. and Redalje D.J., 2006, "CO<sub>2</sub> mitigation and renewable oil from photosynthetic microbes: a new appraisal," *Mitigation and Adaptation Strategies for Global Change*, 2006.
- 38 Photoautotrophic algae use photosynthesis to convert light energy into chemical energy. Some open systems are incorporated into aquaculture systems or as a means of water and waste treatment for existing municipal wastewater treatment systems. Under these conditions, systems may cultivate an indigenous and heterogeneous polyculture of phototrophic algae.
- 39 A pH of approximately 7.0 for freshwater or 8.0 for seawater are typical parameters for algae growth (Huntley and Redalje, 2006).
- 40 Thus, *Chlorella* grows well in nutrient-rich media, *Spirulina* requires a high pH and bicarbonate concentration, and *Dunaliella salina* grows at very high salinity. Personal communication with Jennifer Holmgren, Renewable Energy and Chemicals Division Director, UOP (December 13, 2008).
- 41 Research tells us that viruses or virus-like particles (VLPs, which occur in very few eukaryotic algae and only during certain stages of algal growth), reported in at least 44 taxa of freshwater and marine eukaryotic algae since the 1970s, tend to be strain specific. The structure and composition of these viruses protect them from exposure to organic solvents, which will impact their infectivity. Viruses may be expressed as either symbiotic (advantageous, e.g., contributing to nutrient cycling), neutral, or lethal, and may even contribute to the termination of algal blooms (van Etten J.L. and Graves M.V., 2008, "Phycodnaviruses," *Encyclopedia of Virology, Third Edition*, 4:116-125).
- 42 Personal communication with Al Darzins, NREL, December 13, 2008.
- 43 Personal communication with Jack Lewnard, VP/CTO, Gas Technology Institute, October 13, 2008; Sheehan J., Dunahay T., Benemann J.R. and Roesler P., 1998. A look back at the U.S. Department of Energy's aquatic species program: biodiesel from algae, Golden, CO, National Renewable Energy Institute, NREL/TP-580-24190:113.
- 44 Mockus S.M., Sciasy D. and Johnson R., 2009. "Algae for biofuels: current potential for commercial development." Webinar. William Bardosh (host). The Garbrook Knowledge Resource, Advanced Biofuels Resource (March 11, 2009); GSPI, 2008. Green Star Products complete algae demonstration report.
- 45 Personal communication with Jack Lewnard, VP/CTO, Gas Technology Institute, May 14, 2008.
- 46 Species able to grow in highly saline or high pH environment are known as extremophiles.
- 47 The NREL ASP (1996) sponsored the 1985 SERI Resource Evaluation Report, assessing the availability and suitability of land and water for algae cultivation in the American Southwest, Maxwell E.L., Folger A.G. and Hogg S.E., 1985, Resource evaluation and site selection for microalgae production systems, *Report*, Solar Energy Research Institute, Golden, Colorado, SERI/TR-215-2484.
- 48 A resource assessment, conducted by the Solar Energy Research Institute (SERI) and commissioned by the ASP, concluded that the cost of water lifting (water supply) via pipeline to supply a 400-hectare open pond cultivation system in the arid Southwest would be equivalent to the total cost of financing the construction and operation of the cultivation system. The 1982 ASP report estimated an expenditure of approximately \$31 million. Such a cost would not likely make a feasible business plan (Vigon B.W., Arthur M.F., Taft L.G., Wagner C.K., Lipinsky E.S. et al., 1982, "Resource assessment for microalgal/emergent aquatic biomass in the arid southwest," *Battelle Columbus Laboratory Report*, Solar Energy Research Institute, Golden, Colorado).

- 49 While PVC and polyethylene are less expensive synthetic liner materials, for example, they are prone to UV degradation and stress cracking and may therefore have a shorter lifespan. HPDE and EPDM are more durable but more expensive membranes, and typically have a 20-year warranty.
- 50 Regionally specific data measured by the United States National Weather Service, using the four-foot diameter Class A evaporation pan.
- 51 Carbon sequestration could be an important use in the right situation. In most waters, it is an economic and environmental benefit rather than a requirement.
- 52 Personal communication with Arthur Grossman, Carnegie Institution of Washington, December 17, 2008.
- 53 Variations on these PBRs can be characterized by their shape or configuration, such as helical, spiral, serpentine, parallel, or inclined.
- 54 Ugwu C.U., Aoyagi H. and Uchiyama H., 2008. "Photobioreactors for mass cultivation of algae." *Bioresource Technology* 99:4021–4028.
- 55 Polyethylene bags typically operate in batch mode. Large-scale tubular and flat-plate PBR systems operate in continuous or semicontinuous mode for about two weeks at a time before undergoing routine cleaning and maintenance. Whereas the culture mode is how the system operates, culture densities and percent of lipid biomass produced will determine system efficiency. Batch (also fed-batch) cultures must be harvested in total and replaced with another culture. Batch cultures are known to have a hydraulic retention time (HRT) of up to 28 days. With a constant need for maintenance, batch cultures may be most applicable to smaller systems (e.g., see Appendix B for more on culture modes). At optimum culture density, semicontinuous cultures are partially harvested, leaving behind enough to inoculate fresh culture medium. Semicontinuous cultures have a hydraulic retention time (HRT) of 2–7 days. Continuous cultures have an HRT of 1–3 days. The time period between cleaning and maintenance may differ depending on the desired output. Systems for cultivating algae for food or pharmaceutical products will require periodic cleaning to ensure quality. Although pharmaceutical products are not coproducts of biofuel production, this could still be a limiting factor when cultivation for high-value coproducts requires different cleaning schedules than cultivation for biofuel production.
- 56 Common algal species cultivated in closed PBRs (but not necessarily for energy production) include *Chlorella* spp., *Cryptocodinium colnii*, *Haematococcus pluvialis*, *Synechocystis aquatilis*, *Nannochloropsis* sp., and *Phaeodactylum tricorutum*.
- 57 Mockus, Sciasky and Johnson, 2009.
- 58 The "flashing light effect," helix-configured lighting, and fiber optics are just some of the approaches to delivering artificial light. Optical fibers have been extensively explored in Japan as a potential means of diffusing natural light throughout the culture medium to increase productivity in high concentrations (Muñoz R. and Guieysse B., 2006. "Algal–bacterial processes for the treatment of hazardous contaminants: a review." *Water Research* 40:2799–2815); however, the optical fibers proved costly. Oak Ridge National Laboratory and Ohio University are running a feasibility demonstration for the use of remote solar lighting systems to enhance sunlight utilization and biomass production in PBRs. Large solar collectors on the roof track the sun, collect sunlight, and distribute it through large optical fibers to the bioreactor's growth chamber. The fibers function as distributed light sources to illuminate the algae (cyanobacteria). Alternatively, a multicompart ment PBR, as suggested by Grobelaar and Kurano (Grobelaar J.U. and Kurano N., 2003. "Use of photoacclimation in the design of a novel photobioreactor." *Journal of Applied Phycology* 15:121–126.), allows for differing light intensities to be optimally utilized, subsequently achieving significantly higher productivities when compared to PBRs supplying light with uniform intensity.
- 59 Carbon is commonly pumped into the algal culture in the form of CO<sub>2</sub>-enriched bubbles from a power plant or generator via bubble column. The dissolving rate of gaseous CO<sub>2</sub> may depend on bubble size and the state of saturation of the culture for CO<sub>2</sub>, as well as the pressure and temperature of the reactor (Suh S. and Lee C., 2003. "Photobioreactor engineering: design and performance." *Biotechnology and Bioprocess Engineering* 8:313–321.); Personal communication with David McLaughlin, World Wildlife Fund, October 13, 2008.
- 60 When a culture becomes dense and gas exchange is not maintained, respiration can exceed photosynthesis, which leads to the production of undesirable fermentation acids (i.e., anaerobic fermentation). Therefore, maintaining gas exchange is an important aspect of cultivation whether dealing with a pond or any type of closed bioreactor (Mockus, Sciasky and Johnson, 2009).
- 61 Ugwu C.U., Aoyagi H. and Uchiyama H., 2008. "Photobioreactors for mass cultivation of algae." *Bioresource Technology* 99:4021–4028.
- 62 In 2008, Solazyme Inc. of South San Francisco, California, publicized their successful production of Soladiesel™ in heterotrophically conditioned stainless steel tanks. Although other companies have heterotrophically grown algae for oils (for purified nutraceuticals), Solazyme's product is the first known biofuel produced using a heterotrophic algae-based production system. Amyris and LS9 are companies that have also produced biofuel by heterotrophic fermentation, using bacterial fermentations and not algae.
- 63 Ten hours of fluorescent lamp illumination followed by 14 hours of dark incubation supports continuous growth. In an example given by Miao and Wu (Miao X. and Wu Q., 2006. "Biodiesel production from heterotrophic microalgal oil." *Bioresource Technology* 97:841–846.), 10 g/l glucose was added to the base medium of a cyclic culture. It should be noted that too much organic carbon will lead to mixotrophic growth (leading to more byproducts) when returning to the next light illumination period. During illumination in cyclic systems, inorganic carbon is also pumped into the culture in the form of air enriched with CO<sub>2</sub>. During dark incubation, either ordinary air or CO<sub>2</sub>-enriched air is pumped into the culture (Ogbonna J.C. and Tanaka H., 1998. "Cyclic autotrophic/heterotrophic cultivation of photosynthetic cells: methods of achieving continuous cell growth under light/dark cycles." *Bioresource Technology* 65:65–72:66,69). It is not clear whether cyclic systems are a more sustainable approach to boosting productivity than the continuous lighting of closed PBRs or the continuous glucose inputs of heterotrophic systems.
- 64 Zaslavskaja L.A., Lippmeier J.C., Shih C., Ehrhardt D., Grossman A.R. and Apt K.E., 2001, Trophic conversion of an obligate photoautotrophic organism through metabolic engineering." *Science* 292(5524):2073–2075; Suh and Lee, 2003:317.

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- 66 Personal communication with Arthur Grossman, Carnegie Institution of Washington, October 13, 2008.
- 67 Separating biomass from the water-based culture medium can be an energy-intensive process unless the algae auto-flocculate or settle spontaneously. Personal communication with Jack Lewnard, VP/CTO, Gas Technology Institute, October 13, 2008.
- 68 Personal communication with Matthew Frome and Harrison Dillon, Solazyme, Inc., December, 2008.
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- 70 van Harmelen and Oonk, 2006.
- 71 Gál D., Pekár F., Kerepeczki E., Váradi L., 2007, “Experiments on the operation of a combined aquaculture-algae system,” *Aquaculture International* 15(3–4):173–180.
- 72 The ATS screens on which the algae grow mimic an ecological process in an engineered environment (as opposed to engineering the algae). The flowway slope typically ranges from 0.5–2.0 percent and is largely a function of terrain, cost of grading, and the available algae community.
- 73 Personal communication with Walter Adey, Curator of Algae, Smithsonian Institute, December 2008; Gál et al., 2007.
- 74 Harvesting (once a week) the algal biomass from the screens (by draining, sun-drying, scraping, and vacuuming) mimics the grazing of fish, snails, and other herbivorous aquatic life (personal communication with Walter Adey, Curator of Algae, Smithsonian Institute).
- 75 Shilton A.N., Mara D.D., Craggs R. and Powell N., 2008, “Solar-powered aeration and disinfection, anaerobic co-digestion, biological CO<sub>2</sub> scrubbing and biofuel production: the energy and carbon management opportunities of waste stabilisation ponds,” *Water Science and Technology* 58(1):253–258.
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- 78 Freshwater periphyton are microalgae, while marine (turf-forming) periphyton tend to be macroalgae (Graham and Wilcox, 2000).
- 79 Graham L. and Wilcox L.W., 2000. *Algae*. Prentice Hall, Inc.: Upper Saddle River, NJ.
- 80 Fitoplancton Marino, Producción de microalgas: Acuicultura, accessed December 22, 2008, <http://www.easyalgae.com/aquaculture.asp>.
- 81 ATS system testing indicates the potential to yield 3,500 gallons per acre each year of a mixture of biodiesel and butanol. Volume for volume, this is 15–20 times more than the ethyl alcohol produced from an acre of corn. Adey W.H., 2008, ATS–Energy Project, Progress Report to the Lewis Foundation, p.4.
- 82 Advanced, integrated algal-bacterial wastewater treatment systems have been in operation in St. Helena and Hollister, California for more than 20 years. Though not intended as energy production facilities, these two small-scale systems prove that the feasibility and long-term value of biological-based systems can equal or exceed conventional wastewater treatment. The St. Helena and Hollister wastewater treatment plants produce methane gas as an energy byproduct (Lembi C.A. and Waaland J.R., 1988. *Algae and human affairs*. Cambridge University Press: Cambridge:258).
- 83 Oswald W.J., 2003. “My sixty years in applied algology.” *Journal of Applied Phycology* 15:99–106.
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- 85 Oswald, 1990.
- 86 AIWPS are known to efficiently remove up to 1,000 mg/L of BOD<sub>5</sub> concentration (Ibid.)
- 87 van Harmelen and Oonk, 2006.
- 88 The eutrophic water flows to an HRAP for photosynthetic oxygenation of the settled sewage; this step allows the algae to remove nutrients and release oxygen directly into the water. Wastewater flows into a facultative pond, where sewage solids settle. All the organic solids are anaerobically digested and converted to biogas, almost eliminating sludge accumulation. Oswald, 2003:100.
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- 90 Personal communication with Walter Adey, Curator of Algae, Smithsonian Institute; Oswald, 2003:99.
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- 102 Although Nurdogan and Oswald (Nurdogan Y, and Oswald W.J., 1995, “Enhanced nutrient removal in high-rate ponds,” *Water Science and Technology* 31(12): 33–43) argue that this approach can often be incomplete when calcium and magnesium concentrations are insufficient, research conducted by Lee et al. (Lee S.J., Kim S.B., Kim J.E., Kwon G.S., Yoon B.D. and Oh H.M., 1998. “Effects of harvesting method and growth stage on the flocculation of the green alga *Botryococcus braunii*.” *Letters in Applied Microbiology* 27(1):14–18) illustrates that induced alkalinity (to pH 11) of *Botryococcus braunii* was the most effective method of flocculation.
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- 113 Personal communication with Arthur Grossman, Carnegie Institution of Washington, October 13, 2008.
- 114 Ibid.
- 115 Global Green Solutions, Inc., uses supercritical fluid extraction for the production of vegetable oil (for conversion to biodiesel fuel) and considers it the most environmentally friendly algal oil extraction method available.
- 116 For more on this technology, see OriginOil’s website, <http://www.originoil.com/technology/quantum-fracturing.html>.
- 117 Personal communication with Evan Smith, Co-Founder/ Partner, Verno Systems, June 3, 2009.
- 118 Personal communication with G.S. Heffelfinger, SNL Biofuels Program Lead, Biological and Energy Sciences Center, DOE, August 21, 2008.
- 119 In 2003, there was a lethal hexane gas explosion at a soybean processing plant (producing soybean oil and soybean meal) in Sergeant Bluff, Iowa. Combustible gas detectors could help prevent such disasters, but not necessarily in the extraction plant environment where the detectors are exposed to high temperatures and other inhibitors such as moisture and oil (MSA Instrument Division, [www.msagasdetection.com](http://www.msagasdetection.com)).
- 120 Personal communication with Grant S. Heffelfinger, SNL Biofuels Program Lead, Biological and Energy Sciences Center, DOE, August 21, 2008.
- 121 Personal communication with Walter Adey, Curator of Algae, Smithsonian Institute, December, 2008.
- 122 For a more in depth non-technical overview of conversion technologies, see NREL’s Biomass Research website, <http://www.nrel.gov/biomass/projects.html>.
- 123 Biopolymers are a class of large molecules (polymers), composed of repeating structural units, produced by living organisms (e.g., microalgae). Cellulose and starch, proteins and peptides, and DNA and RNA are all examples of biopolymers, in which the monomeric units, respectively, are sugars, amino acids, and nucleotides. Residual biomass from oil extraction or conversion processes is often comprised of biopolymers that can be converted to animal feed or other high-value products.
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- 141 Personal communication with Jennifer Holmgren, Renewable Energy and Chemicals Division Director, UOP, December 15, 2008; Minowa T. and Sawayama S., 1999. "A novel microalgal system for energy production with nitrogen cycling." *Fuel* 78:1213-1215. ; Patil, Tran and Giselrød, 2008. Sawayama S., Inoue S., Dote Y. and Yokoyama S-Y., 1995. "CO<sub>2</sub> fixation and oil production through microalga." *Energy Conversion and Management* 36(6-9):729-731; Tsukahara, 2005.
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- 150 It may be of interest to note that some species of anaerobic bacteria, including members of the genus *Clostridium*, can convert sugars to acetic acid directly without using ethanol as an intermediate.
- 151 EPA Integrated Risk Information System (IRIS), Acetone (CASRN 67-64-1), <http://www.epa.gov/ncea/iris/subst/0128.htm>.
- 152 Agency for Toxic Substances and Disease Registry, <http://www.atsdr.cdc.gov/tfacts21.pdf>.
- 153 Data Safety Sheet: Acetone, <http://www.jmloveridge.com/cosh/Acetone.pdf>.
- 154 Personal communication with Arthur Grossman, Carnegie Institution of Washington, December 18, 2008.
- 155 The term "tar" lacks a broadly accepted definition in the field of biomass gasification, however, the complex mixture of heavy and aromatic hydrocarbon is said to include those with a molecular weight of 78 (benzene) or higher (Dayton, 2002).

- 156 The low-tar BIG gasification system was developed in Denmark for the medium- to large-scale power plants (2–20MWe) and is designed to produce gas with little or cheap need for unit cleaning. The gasification process is based on use of moist fuels (40–60 percent water content); however, dry fuels can also be used. The low-tar, high-efficiency biomass gasification concept is referred to as “low-tar BIG” (Biomass Integrated Gasification). The concept is based on separate pyrolysis and gasification units. The volatile gases from the pyrolysis (containing tar) are partially oxidized in a separate chamber, thereby dramatically reducing the tar content. According to Anderson et al. (2008) the investment and running cost of a gas cleaning system can be reduced and the reliability can be increased (Anderson L., Elmegaard B., Qvale B., Henriksen U., Bentzen J.D. and Hummelshøj R., 2008, “Modelling the low-tar BIG gasification concept,” Joint paper from the Technical University of Denmark and COWI).
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- 192 The USDA-APHIS was given authority to monitor field trials of new genetically engineered (GE) varieties and also to “deregulate” GE crops for commercial cultivation based on the Federal Plant Pest Act (1957) and the Federal Plant Quarantine Act (1912). In 2000, the Plant Pest Act was superseded by the Plant Protection Act, which also does not specifically address the genetic modification of plants.
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